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EDITED BY

PAUL C. FREER, M. D., PH. D.

AND

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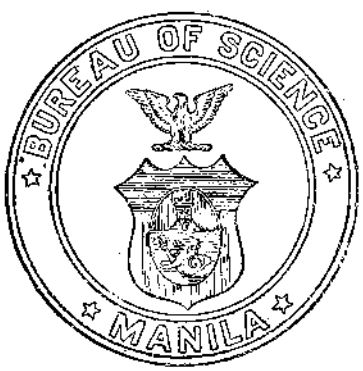

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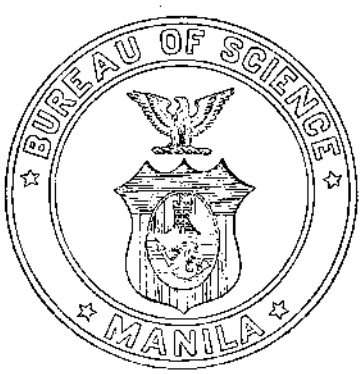
WITH 34 PLATES, 5 FIGURES, 11 CHARTS AND 1 DIAGRAM



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THE PHILIPPINE

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B. MEDICAL SCIENCES

VOL. V FEBRUARY, 1910 No. 1

THE TROPICAL SUNLIGHT.[1]

By PAUL C. FREER.

(From the Bureau of Science, Manila, P. I.)

INTRODUCTION.

I have chosen my topic for discussion, not only because I knew that an audience, every one of whom has had a more or less extended experience in the Tropics, would be vitally interested in it, but also because in the past decade differences, more especially of insolation, in portions of the globe showing contrasts in climate have been the subject of extended monographs and papers appearing in scientific publications and journals. For the greater part, this literature has especially to do with the objective manifestations of the effects of equable temperatures, possible humidity, and supposedly intense sunlight upon the conditions of life of human beings and their response to their environment. Considered in this aspect, there always enters an element of uncertainty owing to the absence of absolute means of measurement and the variability of other hygienic surroundings. The people of the Tropics, by reason of their mode of life, their food, their backwardness in scientific procedures and their superstitions, are exposed to many infections and causes of disease which modify any conclusions which may be drawn, and these effects, because of their omnipresence, reflect themselves upon the modern European intruders. Experiments

[1] Address of the president at the first biennial meeting of the Far Eastern Association of Tropical Medicine, held at Manila, March 5, 1910.

conducted upon the influence of this or that type of clothing, on the modifications of covering upon troops in the field and other similar lines of investigation always suffer from the factor of the condition of the subjects themselves. The impossibility of introducing any exact comparisons into these experiments renders the conclusions drawn therefrom arbitrary.

However, in almost all cases, differences in the environment brought about by contrasts between tropical and temperate climates are referred in the first instance to the sunlight, in the second to humidity, if this factor is taken into consideration at all, and in the third to the equable temperature throughout the year; that is, to the absence of a pronounced winter.

It has seemed to me that the first of these factors, the sunlight, is capable of some exact comparative measurements, although, from the nature of the case, the experimental difficulties in the way are very great. To draw any final conclusion, work with reliable and calibrated instruments must be undertaken in various parts of the globe, through a long series of days, and experiments must be planned so as to eliminate, so far as possible, the personal errors which may creep in. We must take into consideration the average number of hours of sunshine per day, the degree of haziness or cloudiness, the variation with the seasons, the presence or absence of pronounced winds and the proximity to the seashore of the stations as well as their altitude. Needless to say that, while many individual observations exist, no comprehensive plan of work, based upon data taking cognizance of all of these factors, has ever been carried into effect. In order, if possible, to make a beginning, I have brought together the results of the investigations which we have been able, so far, to conduct in this institution, together with such as may be pertinent to the question obtained in other places. The sum total leaves us with only a few facts gained and a few conclusions to be drawn, but it shows to a certain extent what direction it is best to follow in coöperative work and it also reveals to us the complexity of the field.

THE SUNLIGHT.

We all know that the sunlight can be dissolved by proper instruments, such as a diffraction grating or a prism, into an infinite number of waves or pulsations of different length, which produce a continuous visible spectrum extending from deep red to violet; this spectrum being crossed by numerous sharp lines, representing the absorption phenomena of the elements existing in a gaseous state in the sun. On either side an area not visible exists, the infra-red and the ultra-violet. The latter portions of the spectrum can be studied by means of photographs. Measurements are made in millionths of a millimeter, starting from some well-known line which may be produced between two poles of a given metal by

a spark caused by a current of high tension. If we compare the spectrum of the sun's light with that of a metal, such as cadmium, we discover that the latter shows lines extending into the ultra-violet field far beyond that of the former, yet it is evident, if we consider the elements known to be present in the sun's mass and chromosphere, by a study of the spectrum of the sunlight, that outside our atmosphere, or at least in the central body of the sun, such ultra-violet rays must be present. It follows that the air surrounding our globe must be able to absorb or otherwise modify a large proportion of the latter. Measurement can show whether, as a rule, there is a greater or less absorption in the Tropics than in temperate climates.

The theory was prevalent some years ago that the spectrum was divided into classes of rays, of which those in the blue, violet and ultra-violet were considered as "actinic," namely, as producing chemical reactions, those in the visible field as causing light, and those in the ultra-red as heat, although even as long ago as 1859, Bunsen and Roscoe [2] indicated that no sharp lines could be drawn between the rays which give rise to the thermic phenomena in the atmosphere and on the earth, and those causing chemical changes. The power of seeing is only a subjective symptom, some persons may have a range of vision for light rays beyond others; all rays may produce chemical changes, yet it is true that some of the latter are brought about much more rapidly by the portion of the spectrum lying toward the violet and beyond, than by the other part toward the infra-red; indeed, in many changes the latter can not act as a catalyzer at all. However, we must not lose sight of the fact that the total energy derived from the sun, in which must be included rays of all lengths, is an essential factor in maintaining chemical changes on the earth, regardless of the fact that recent investigations have made us cognizant of other sources of energy which are supplied by the mass of the globe itself. The quantity in which the former reaches the earth's surface at different places may have much to do with the effect of local environment.

In studying the manifestations of energy in other branches of science, we endeavor to divide such manifestations into two factors, one of intensity or stress, and the other of capacity. So, in electricity we have volts (stress) and coulombs (capacity); in the effects of gravity, distance (stress) and mass (capacity). Light can be treated in the same way. Let us suppose we have an area of 1 square centimeter through which rays of a given wave length are passing from a constant source of light. If we double the opening, we would have twice the light passing through, but if we concentrate the total number of rays entering the opening of 2 square centimeters, by means of an ideal lens, on the one we would obviously have doubled the amount of light in the same time passing through the slit, the area of which had not changed, in other words, we

[2] *Ann. d. Phys. u. Chem.* (Poggendorf) (1859), **108**, 193.

would have increased the intensity of illumination, or the amplitude of the light waves, a process corresponding to an increase of voltage, with diminution of the diameter of the cross section of the wire, in an electric conductor. While it is easy to conceive of measurements of this kind where rays pass through a definite, limited opening, it is not so easy to see how they can be made when the illumination extends over a vast area. Bunsen and Roscoe [3] and Roscoe and Baxendell,[4] in their classical researches on photochemistry, employed an explosive mixture of chlorine and hydrogen to measure the intensity of insolation, and by a variation in the quantity of light, by using different sources and different diameters of openings, came to a series of results which, more nearly than any other, can be used as a basis of calculation. However, it seems to me that the possibilities of autocatalysis in such a mixture,[5] which have since then become understood, would have a great influence in the calculation of the total effect of insolation in climates other than the one in which these investigators worked. Subsequently, they also perfected a method based upon the darkening of a normal strip of sensitized paper in known intervals of time to standard colors, and this means, if the paper were always correct, could be developed in the direction mentioned. Indeed, Roscoe [6] described a self-registering apparatus for use in meteorological stations which would, if universally adopted, already have made available the data sought for. However, it seems to me that all methods which consider "photochemical" light only as a group, without division by the spectrum, are faulty because they do not differentiate the proportion of waves of various lengths contained in the total insolation at different latitudes and under different meteorological conditions. The range of rays which will bring about the union of hydrogen and chlorine, or the darkening of sensitized paper, is very great, whereas it may be only certain very limited portions of the spectral field which produce maximum results.

One method suggests itself. If two spectra of the sun, each showing the same area, that is, composed of waves of the same length, were to be projected upon two extremely slow photographic plates, so slow that, although the plates are approximately the same, slight variations in time would have no appreciable effect upon the total exposure, and if comparisons were to be made as to the time necessary to produce the first visible image of some given line, then the ratios of these times would, for most practical purposes of comparison, give the ratios of intensities. The accuracy would increase by selecting more than one line, successively.

[3] *Loc. cit., ibid.* (1862) **117**, 529.

[4] *Ann. d. Phys. u. Chem.* (Poggendorf) (1866) **128**, 291.

[5] Chapman, Chadwick and Ramsbottom, *Journ. Chem. Soc. London* (1907) **91**, 943, give a parallel case in the reaction of carbon monoxide and oxygen in the presence of moisture. Hydrochloric acid takes the place of the latter.

[6] *Ibid* (1877) **151**, 268.

However, other chemical reactions could be selected, perhaps with less experimental difficulty, provided all side reactions, such as oxidation or peroxide formation, were eliminated. Such a reaction is the decomposition of oxalic acid in the presence of uranium acetate as a catalyzer, the gas evolved in a given time by given rays being measured, or the residual oxalic acid titrated. This reagent for the measurement of the two factors of the sunlight without, however, as yet using definite lines, has been utilized by Dr. Raymond F. Bacon, of the Bureau of Science, he being able to compare the insolation at Chicago, in June, with that of Manila throughout the year. I will discuss his results in another portion of this paper. The method being fully developed, subsequent experiments, using definite lines of the spectrum will be a simple matter.

His measurements, although extending over a limited period in Chicago, are nevertheless of such a nature that we have an experimental comparison between the effect of insolation in the Tropics and at a northern point. Previous attempts have been made to calculate the effect from the data of Bunsen and Roscoe,[7] taking into consideration the direct and diffused sunlight. The calculations result in the development of numbers showing that "diffused light tends to equalize the numbers for the total quantity of light at different latitudes." So, according to Sebeliene,[8] while "the daily quantity of light due to direct radiation is forty times as great at the equator as it is at the pole, the quantity of diffused daylight is hardly twice as great at the equator as at the pole on the same day." The results of Sebeliene's calculations, based on Bunsen and Roscoe's figures, give a total quantity of light units at 0° of latitude as 82,716, a maximum of 114,835 at 30°, and a minimum of 76,048 at the pole. However, these calculations refer only to the midsummer day, and certainly lack the basis of experiment in various parts of the world. While the northern and southern parts of the globe are much more fortunately situated at midsummer in respect to light, as compared with the Tropics, this would not hold good throughout the year. Bacon, in the Bureau of Science, found the decomposition of oxalic acid in the presence of uranium acetate to proceed approximately five times as fast in Manila in October and November as in Chicago during the months of May and June; and recent days in February have shown a rate as high as twenty. Bunsen and Roscoe[9] also demonstrated that days of light, hazy cloud, through which the sun just shines, are able remarkably to increase the chemical activity of light.

Enough has been said to demonstrate the difficulties to be encountered in securing data fit for comparison; the experimental work so far accomplished, therefore, is as yet only tentative and must be taken for what it is worth.

[7] *Loc. cit.* (1859), **108**, 257, 260.
[8] *Phil. Mag.* (1905) (VI), **9**, 354.
[9] *Loc. cit.* (1859), **108**, 236.

THE COLORATION OF PHENOL, OF ANILINE, AND OF METHYL ALCOHOL IN THE SUNLIGHT OF MANILA.

All of us who have experience with laboratories in the Tropics presume, in a general way, that chemical reagents are much more subject to deterioration and change in such regions than in temperate climates, although the containers may be securely sealed from the air. Is this so, or is it simply a hasty generalization, brought about by our personal bias, driving us in the direction of thinking the phenomenon must really exist because of preconceived notions of the influence of tropical environment?

Phenol is eminently fitted for study, because some data for comparison, gathered in other climates, exist. Mr. H. D. Gibbs,[10] of the Bureau of Science, has occupied himself with this subject for more than a year, and I will give a brief summary of his results.

The phenomenon of the coloration of phenol in the sunlight is one of oxidation, it does not occur in an atmosphere of an indifferent gas such as hydrogen or nitrogen. The change takes place even with absolutely pure, dry phenol and dry oxygen, so that the presence of moisture is not necessary. The reaction, under ordinary conditions, therefore, is inaugurated independently of the degree of humidity of the atmosphere, although the water (or hydrogen-peroxide) produced by the beginning reaction will undoubtedly have an accelerating effect on its subsequent rate. Crystals of such pure, dry phenol, when sealed in a tube with pure, dry oxygen and placed in the sunlight in Manila at a temperature of approximately 30°, color perceptibly after two hours and the entire mass changes to a deep red liquid after five days.

This rate of coloration probably varies with the seasons. When the sun is directly overhead there is apparently a much more rapid production of color than in December and January; and it is more rapid under quartz than when the sample is exposed under soda glass. This latter difference would seem to point to the fact that the change is inaugurated by and accelerated in a greater degree by the ultra-violet portion of the spectrum, not absorbed by quartz glass, than by the others, although the effect can be brought about by heat alone, but much more slowly.

The absorption spectum of phenol[11] gives a broad absorption hand at $\lambda=272\ \mu\mu$. There is, therefore, according to Baly and Collie,[12] a condition of unstable equilibrium of one hydrogen atom in each molecule of phenol, at a given temperature in the dark. Therefore there would be a constant ratio between the large proportion of phenol in the enol form and the minute quantity of the keto (quinone) form. The action of light, especially of that of the ultra-violet portion of the spectrum, would therefore be to change the equilibrium by increasing the proportion of the latter present, and hence the rate of oxidation; it being presumed that the enol form, the one ordinarily accepted for phenol, is not attacked by the oxygen. No quinone form seems to be present in the crystals. Anisol is not colored by oxygen or ozone in the presence of sunlight, hence it is to be presumed that no such labile form exists in the latter.

[10] *This Journal, Sec. A* (1908), **3**, 361; *Ibid.* (1909), **4**, 133.
[11] Hartley, *Journ. Chem. Soc.* (1902), **81**, 929.
[12] *Ibid.* (1905), **87**, 1339.

The color change of phenol takes place much less rapidly in temperate climates where measurements have been made. Richardson gives three days in Clifton, England,[13] and in some instances several weeks have not sufficed to produce the color. A tube of phenol and oxygen, in the dark, kept at a temperature of 100° for two weeks, gave only a faint yellow color, and phenol under the same conditions in the diffused light of a laboratory room at Manila at a temperature of approximately 30° was not appreciably colored after two months.

The products of oxidation are quinol, quinone and catechol, all of which confirm the conclusion that the change to the quinone formula is markedly accelerated by the ultra-violet rays which are absorbed by phenol and point in the direction of the supposition that these rays are present in greater proportion in the sunlight of the Tropics, at sea level, than in more northern climates. The conclusion in regard to the labile condition of one-sixth of the hydrogen in phenol would probably also be found true for a large series of similar compounds, and may, possibly in the future be extended so as to explain a great many chemical phenomena prominent in tropical climates.

Aniline is even more fitted than phenol for a study of the effects of insolation, as the changes are brought about with greater rapidity and the oxidation products are present in greater quantity.

When perfectly pure aniline is sealed in a thin glass tube with dry air and exposed to the sunlight in Manila during the months of April, May and August, it darkens and assumes a decided red shade in less than ten minutes. Of course, the temperature, as in the case of phenol, is also an important factor.

The products of the reaction which have been isolated are azobenzene, 2,5-dianilinoquinone, 2,5-dianilinoquinoneanil, and azophenene. The oxidation is therefore in all probability accompanied by one of condensation, and again it seems not unreasonable to refer the rapid accomplishment of this portion of the reaction to the part of the spectrum lying in the violet and beyond.

Pure samples of aniline also became colored upon long standing in the dark. Aniline, like phenol, therefore, has a portion of its hydrogen labile and probably shows a similar equilibrium between the ordinarily accepted formula and the quinoid structure as is presented by phenol. Its absorption spectrum also gives a deep band in the violet.

However, aniline differs from phenol in the fact that in the tropical sunlight it undergoes chemical changes even in the absence of oxygen, that is, when it is placed either *in vacuo* or in the presence of indifferent gases such as hydrogen, nitrogen, or carbon dioxide. At first it is colored yellow and finally it changes to a brilliant red. The work on this subject is not entirely completed. However, it may be stated that no gas is evolved, azophenene is the main substance formed, and ammonia is also

[13] *Journ. Soc. Chem. Ind.* (1893), 12, 415.

present. The sunlight therefore, in this climate, produces a profound decomposition of aniline, oxidation to azophenene and simultaneous reduction to ammonia and probably benzene taking place. No data for comparison with the same reaction as produced by the sunlight of temperate climates are at hand, but it is safe to presume that a change of the kind just mentioned does not take place except with extreme slowness in northern latitudes; at least none of us have ever become aware of it before working in Manila.

Mr. Gibbs has also taken up the study of the oxidation of methyl alcohol in the sunlight and has shown that formaldehyde is readily produced in this climate, although in other, more northern portions of the earth, no reaction at all could be noted under these conditions. Methyl salicylate is also colored in the sunlight of Manila with a fair degree of readiness, the methyl alcohol produced by the hydrolysis of the ester being oxidized to formaldehyde and the latter condensing with the derivatives of salicylic acid to produce a dye.

All of the reactions which have been mentioned are still the subject of investigation, but enough has been brought out to render certain that the tropical sunlight, in fact, produces chemical changes which either take place much more slowly in temperate climates, or indeed do not take place at all.

THE DECOMPOSITION OF OXALIC ACID BY SUNLIGHT IN THE PRESENCE OF URANIUM ACETATE.

In order, if possible, to obtain a quantitative estimate of the effect of the sunlight at this latitude, and to institute comparisons between this effect and that to be observed in the more northern portions of the United States, Dr. Raymond F. Bacon, of the Bureau of Science, has modified the method adopted by Duclaux.[14]

The latter author, subjecting oxalic acid to the action of the sunlight, in the presence of oxygen, in various parts of the globe, encountered the following remarkable facts.

> Two months of uninterrupted observation showed that the solar combustion passes through very different values within twenty-four hours. These changes are sometimes very sudden and exceed, especially those of the thermometer, the barometer, and even those of the average brightness of the day.
>
> So, for example, on a fine day without clouds, there was 34 per cent of the solution of oxalic acid destroyed, while, only five days earlier in the year, on a day with slight cirrus, 35 per cent were burned. A bright day in October showed 12 per cent as against 20 per cent when the sky was cloudy during the entire time of exposure, and even greater contrasts, from 7 to 28 per cent, were observed on days entirely similar to all outward appearances. The maximum decomposition appears to be in the spring.

[14] *Smithson, Contrib. to Knowledge* (1903), **29**, 1034.

Comparisons made in August between St. Pierre le Port, on the Channel, and Helsingfors (latitude 60° 10′) gave as a maximum 44 per cent of decomposition in France and 77 per cent in Finland, between the hours of 8 and 4, a most remarkable result, tending to show that the chemical effect of the sun's rays, in late summer, is greater at the far northern latitude of Hesingfors than in France. In Algeria, during the same months, the maximum was only 14 per cent. According to Duclaux's experiments, therefore, the chemical effects of the sun's light diminishes in the summer months as we proceed from a northern climate toward the equator.

The above results do not hold good in Manila. Here, following the method of Duclaux, Doctor Bacon has found that, using 10 cubic centimeters of $\frac{N}{10}$ oxalic acid, the decomposition of oxalic acid, between the hours of 8 and 4, is almost always practically complete. Thus, the figures for four days are 68, 96, 85, and 100 per cent, respectively, and even on a day over one-half of which was cloudy (January 19), complete oxidation had taken place by 5 o'clock in the afternoon. While there is no doubt as to the results obtained by Duclaux in the dry climate of Algeria, near a great desert, it certainly is true that those reached in Manila are contradictory to the ones which have been quoted, and that there is here a marked increase in the rate of decomposition.

However, the oxidation of oxalic acid in the sunlight suffers from a great defect. Duclaux noticed that a solution when first exposed decomposed very slowly and that the reaction is subsequently rapidly accelerated. We have shown here that this phenomenon depends on the formation of hydrogen peroxide, and that the increasing rate is due to autocatalysis. While the method, therefore, gives a certain measure of comparison, it is not above criticism.

The decomposition of oxalic acid in the presence of uranium acetate is not dependent upon oxidation, and is not accelerated by autocatalysis, and is therefore much better adapted to comparative study.[15] Doctor Bacon also had the good fortune, while in America on leave, to compare this reaction in Chicago with that in Manila. The time in Chicago was during the months of May and June; the total average extending over ten days gave, for Chicago, approximately 100 cubic centimeters of gas in two hundred minutes, and in Manila, with the same solutions, 100 cubic centimeters in forty minutes. It is also at present noticeable, in this climate, that as the altitude of the sun is increasing, the decomposition is accelerated; thus, January, 1910, gave an average decomposition of 0.081 gram of oxalic acid per hour as compared with 0.090 in the middle of February. The optimum day in Manila has far exceeded the ratio of 5 to 1 observed above, and has more nearly reached 20 to 1.

[15] Raymond F. Bacon. *This Journal, Sec. A* (1907), 2, 129.

ACTINIC AND NONACTINIC DAYS.

The differences between various days apparently similar in every respect as regards insolation is very remarkable, and what is still more striking, hazy days seem often to bring about greater decomposition of the oxalic acid than those which are perfectly clear; conditions which had previously been noted by other authors referred to in this paper.

A table will make this more evident.

TABLE I.—*Decomposition of oxalic acid in the presence of uranium acetate.*

No.	1910.	Decomposition, grams of oxalic acid.	Day.
1	Jan. 17	0.084	Bright sun.
2	Jan. 18	0.083	Hazy.
3	Jan. 19	0.049	Partly cloudy.
4	Feb. 4	0.080	Partly cloudy, but on the whole fair.
5	Feb. 5	0.050	Bright sunlight.
6	Feb. 7	0.085	Sun intermittent but still partly bright.
7	Feb. 8	0.045	Hazy and cloudy.
8	Feb. 9	0.075	Hazy and cloudy.
9	Feb. 14	1.000	Bright sunlight.
10	Feb. 16	0.085	Partly cloudy.
11	Feb. 17	1.120	Bright sunlight.
12	Feb. 18	0.041	Cloudy.
13	Feb. 21	0.047	Cloudy.
14	Feb. 22	0.073	Bright sunlight.
15	Feb. 23	1.500	Bright sunlight.

It is feasible, therefore, to divide even the days of bright sunshine into those which are "actinic" and others which are "nonactinic." Thus, numbers 9, 11, and 15 are distinctly actinic, whereas number 5 and 14 are nonactinic; undoubtedly the effect of insolation in the Tropics during the former would be much greater than during the latter; indeed, some of the nonactinic days are very nearly like those of temperate climates.

THE EFFECT OF TROPICAL SUNLIGHT ON THE ATMOSPHERE.

Another phenomenon to be observed in Manila in a marked degree, and which, so far as I am aware, has not been recorded in the literature from other climates, is the extensive ionization of the air when exposed to the sunlight. Doctor Bacon, using a modern electroscope, has been able to show that our atmosphere, when exposed to the direct rays of the sun, rapidly discharges the instrument, the loss of potential being 46 volts per hour, whereas, in the diffused light of a room it is only 15, and during the night 6, for the same volume of air. This is certainly a remarkable result, which deserves further study. The only comparative data on hand

are a few by Elster and Geitel [16] giving us an indication of what the fall of voltage would be in northern climates. They found, in Vienna on a foggy day, a voltage of 2.77, in clear weather, 8.58, but on a day when the sky was half overcast, 13.67. These authors ascribe the phenomenon to radio-activity, but our results in Manila, where radio-active phenomena are not especially prominent, would lead to the conclusion that the air is ionized by sunlight. The presence of this ionization in so great a degree in our atmosphere would indicate a condition of the solar spectrum which might well account for many of the so-called excessive effects which have been observed.

THE EFFECT OF SUNLIGHT ON MICROÖRGANISMS.

A great mass of literature exists on this subject, but the later publications of Hertel [17] have brought the study down to such a clear basis that it seems scarcely necessary to enter into the work of previous authors with any detail. Suffice it to say, it is a fairly well established fact that bacteria and even their spores are destroyed by sufficient exposure to the sunlight. Of course, the time necessary for this effect varies greatly with the latitude and the degree of insolation, and naturally the errors of observation are very large.

The general method of procedure has been to expose plate cultures or tubes to the direct action of the sunlight during fixed hours, placing controls in the incubator, and covering either portions of the plates or tubes, or their contents where needed, by tin foil. Of course, in the majority of published researches, care was taken to exclude heat action by proper precautions. Different colors were produced by colored plates or solutions, in some instances spectroscopically examined. But few of the authors have used quartz prisms and lenses to give a spectrum. It is well known that glass absorbs the ultra-violet of the spectrum.

Hertel undertook his experimental work not by employing the entire sunlight, or by blending with colored glasses, but by the spectrum of the sun from the ultra-violet region, using an apparatus the parts of which were constructed of quartz, testing the action of the light within different, but sharply defined regions of determined wave length. The first source of light employed was the bright line from magnesium at $\lambda = 280\ \mu\mu$.

Bacteria and vibrios (*B. coli*, *B. prodigiosus*, the vibrio of cholera, *B. typhosus* and others) in the first moments of illumination by this portion of the spectrum, increased in motility, that is, gave evidence of stimulation, but after a few seconds there was a retardation and finally complete rest. The bacteria of decay also were markedly affected, with final death. Especial experiments demonstrated that the light had no effect on the culture media, so that the sterilization resulted because of the action of the radiant energy on the organisms alone.

[16] *Ann. d. Phys.* (IV), **2**, 425. The authors used an instrument of identical form with our own.

[17] *Ztschr. f. allg. Physiol.* (1904), **4**; (1905), **5**; (1906), **6**; *Rev. in Biol. Centralbl.* (1907), **27**, 510.

Paramecia, when exposed, showed an instantaneous state of unrest, with a tendency to escape from the illuminated field, longer illumination finally brought about their complete decomposition. Nematodes were more resistant, death following only after two or three minutes. Larvæ of amphibia showed a decided wandering of pigment toward the spot in contact with the light. All of the organisms investigated had a marked reaction toward light of 280 $\mu\mu$, first in increased motility or contraction of contractile tissues, followed by lassitude, slowness of motion and final death. Toxins and ferments were also tested, so, for example, the toxin of diphtheria was rendered entirely inert after five minutes; trypsin, diastase, and the ferment of rennet were also profoundly affected, but not in as great a degree as the toxins.

The experiments next were extended to lights of various wave lengths, the total energy of each region of the spectrum being measured thermo-electrically, it developing that *"the action of one and the same spectral line is directly proportional to its total intensity, as measured thermo-electrically."*

The physiological action of the rays diminishes with increasing wave length, a difference of 50 $\mu\mu$ showing a marked effect. Light of 280 $\mu\mu$ kills organisms almost at once, whereas that of 440 $\mu\mu$, of equal intensity, does so only after a number of hours of action.

Perhaps the most important result of Hertel's work, for our present purposes, is his proof that not only the total energy and the wave length of the incident light are of importance, but also the relative proportion of the rays absorbed by the organisms, for he developed the fact that the absorption of radiant energy by the tissues of the organisms investigated is diminished the longer the wave length of the light which is employed.

The action of a specific kind of light on organisms is therefore not only dependent on its total intensity, but is also in the greatest degree related to the power of absorption for this light possessed by the tissues. By means of erythrosin Hertel was able so to stain the cells of living organisms that their power of absorption for waves of greater length than 280 $\mu\mu$ was markedly increased. The introduction of this experimental modification enabled Hertel to demonstrate that even the visible light rays could bring about destruction of tissue in the same time as the ultra-violet.

The physiological action of light rays is therefore not dependent upon any specific region of the sun's spectrum, the wave length is only of importance in such degree as the total energy, and the power of absorption is determined thereby. A plant, for instance, for the existence of which light is absolutely necessary, takes on a color which is complementary to the incident rays, for example, *Oscillaria sancta* colors red in green light and green in red. Light also has an unfavorable effect on the phenomenon of cell division, but this only takes place with higher intensities. Different pigment cells have a different absorptive power for incident light, according to its wave length, but the ultra-violet rays are equally absorbed by all. Therefore, the latter differ markedly from visible light, in which the absorption maxima, according to the color, lie at diverse and far-removed parts of the spectrum.

The character of the pigment, therefore, is of fundamental importance in determining the effect of insolation for all save the ultra-violet rays; but the experiments of Baly on chemical substances, in distinction to

those of Hertel on the pigments of natural tissue, have shown that the absorption of the former for ultra violet light differs markedly according to the nature of the chemical individual employed.

While Hertel's investigations have no direct bearing, at present, on the subject of tropical sunlight, because concrete and connected experiments in the Tropics are lacking, still the coloration of phenol and aniline, the oxidation of methyl alcohol, the rapid decomposition of oxalic acid in the presence of uranium acetate, and the intense ionization of the air in Manila, are possibly referable to a larger proportion of ultra-violet light in the sun's spectrum at this latitude; and therefore, if this be so, it will also be true that living tissues will be affected abnormally, or, in other words, we should expect the effects of tropical sunlight upon microorganisms and those of a higher order, and the consequent endeavor by pigmentation or other means of these organisms to protect themselves, to be similar to those observed by Hertel for the ultra-violet in his spectroscopic investigations. Of course, we must not lose sight of the fact that the tropical sunlight probably displays a greater intensity of all rays, and that this effect would be cumulative.

Ewart[18] calls attention to the fact that plants in the sunlight of the Tropics seek to protect themselves by the production of a red color (erythrophyll) the absorption spectrum of which has a band in the violet. In a journey to Java he observed that the red color of the leaves was more common and marked in the low, tropical valleys than higher up on the mountains. In the cloud-covered belt it was entirely absent, but above the latter it again appeared.

However, it has not been proved that the tropical sunlight really does contain a greater proportion of ultra-violet light than that of northern climates. So far, we can only come to the conclusion that it has a greater intensity, as the experimental work which I have given has shown, and that a certain proportion of rays lying in the ultra-violet is present. Greater intensity, although with but a small area in the latter portion of the spectrum, would bring about results similar to those which would be observed if the light extended to 280 $\mu\mu$ beyond.

THE SPECTRUM OF THE SUN IN MANILA DURING FEBRUARY, 1910.

We have endeavored to solve this problem in Manila by the construction of an instrument from such means as are at hand. Funds have heretofore been lacking to have made by the best mechanics of America or Europe a spectro-photographic apparatus entirely adapted to the needs of the investigation.

Our spectroscope consists of a Rowland grating, a heliostat, both very kindly loaned to us by the Philippine Weather Bureau,[19] and a quartz lens and slit taken

[18] *Journ. Linn. Soc. (Bot.)* (1895), **31**, 364; *Ann. Bot.* (1897), **11**, 440.

[19] I wish at this place to express my sincere thanks to the Reverend Father José Algué, Director of the Weather Bureau, at Manila, for his loan of apparatus and his most kindly coöperation in all that we have asked of him.

from a Zeiss ultra-violet photomicrographic apparatus. The three are mounted in a line upon a prism base, and the image of the spectrum is projected upon a camera placed at one side, and in the plane of the slit. The necessary motions of camera, lens and grating are provided, but, unfortunately, we can not work with curved plates, so that only a small portion of the spectrum is in focus at one time. The whole apparatus is inclosed in a light-tight box and secondary spectra are excluded by an appropriate shield within the box. (See fig. 1.) The use of the heliostat can be avoided by pointing the apparatus directly at the sun.

The spectra obtained at noon are shown by Plate I. They do not probably extend beyond $\lambda=291\ \mu\ \mu$, and therefore not much farther than has been observed by others. Measurements undertaken by Miethe and Lehmann [20] in Assuan, Berlin, Zermatt, Gornergrat, and Monte Rosa give practically identical numbers, namely 291.55 $\mu\mu$ to 291.21 $\mu\mu$ during the latter part of August and the first part of September; these authors finding, in contradistinction to the observations of Cornu,[21] that altitude above the sea level makes no great difference. As one of these places is at 24° 30′ north latitude, where the others are in northern climates, it is evident that as the extent of the ultra-violet field does not change materially, the intensity factor in the solar spectrum must vary to a great extent in different places. However, it is possible that a considerable range of ultra-violet is absent at present (March 1) from our sunlight. Probably this area will increase as the angle of the sun diminishes and as the season advances; and it may reach a maximum in April, although these recent results would seem to indicate that even here we will not get below 288 $\mu\mu$.

Enough has already been demonstrated to prove that the emphasis placed upon the ultra-violet spectrum in the preceding part of this paper is justified.

GENERAL CONSIDERATIONS.

If we consider the equator as being in one plane and the poles in another, normal to the incident rays of the sun, it is obvious that the relative distance between these two points is practically infinitely small as compared with the distance of the sun from the earth. If we regard this phase alone, obviously the insolation at the equator and the poles would approximately be equal. However, these two positions are on different points of a sphere, which is surrounded by the atmosphere, so that not only is the angle of incidence of light rays different in different regions, but also, as they pass from the ether into the atmosphere at different angles, they suffer refraction in differing degree. The shorter wave lengths, in this respect, are influenced to a greater extent than the longer, and, under proper conditions, are even finally totally absorbed or reflected, so that none would reach the earth. This condition would

[20] *Sitzungsber. d. k. Pr. Akad. d. Wissensch.* (1909), **8**, 268.
[21] *Compt. rend. Acad. Sci.* (1879), **88**, 1107; *ibid* **89**, 808.

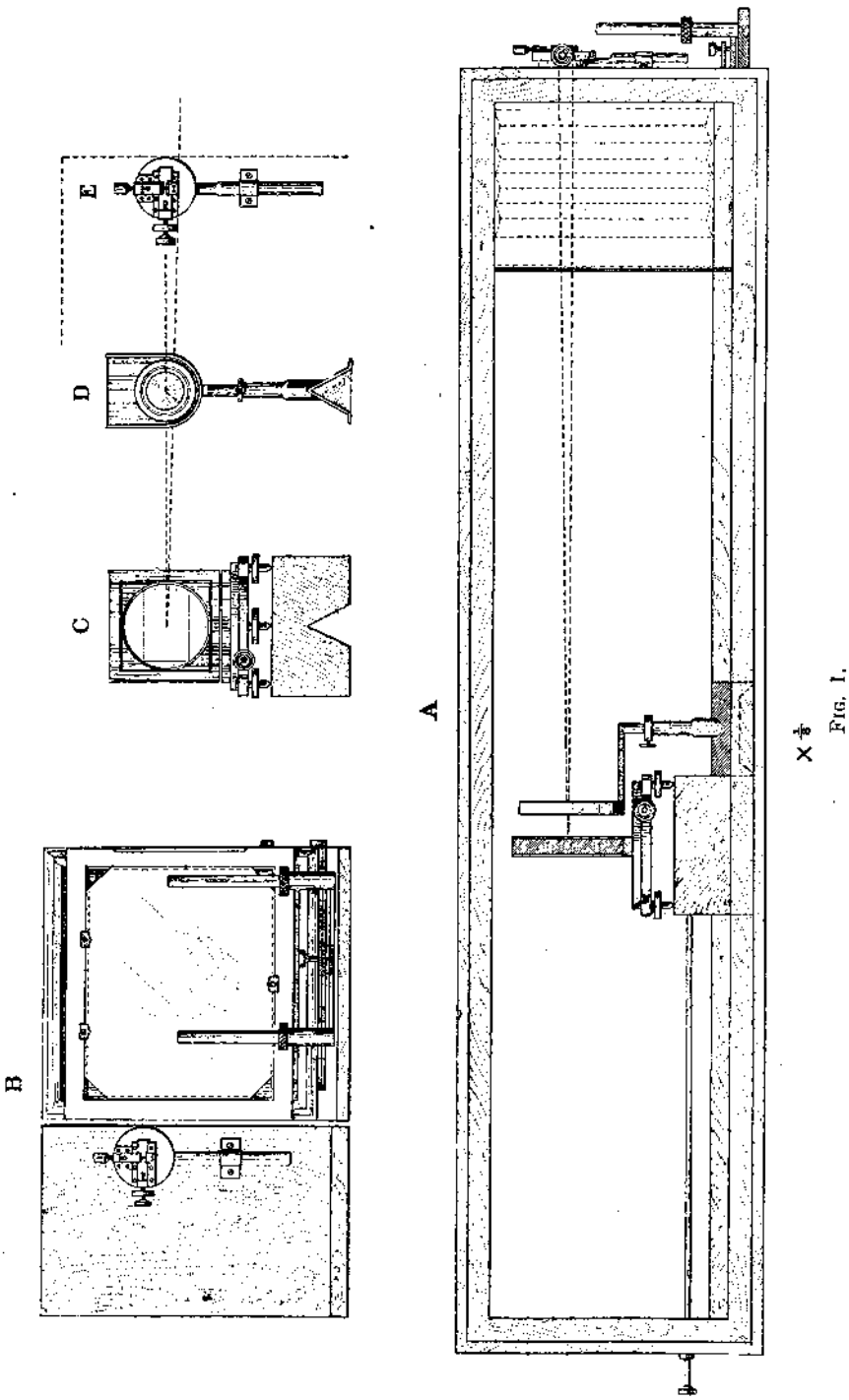

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FIG. I.

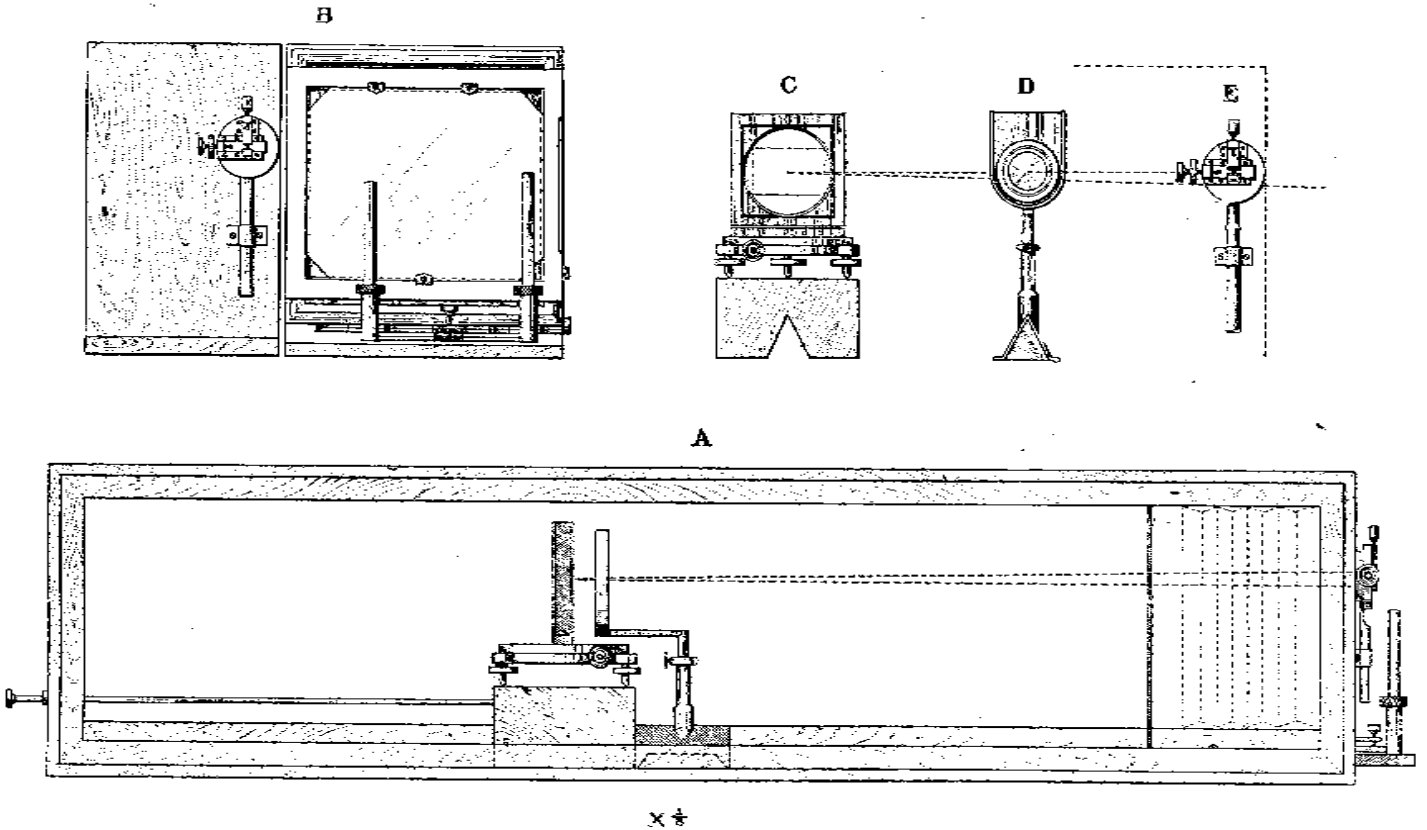

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FIG. 1.

come about the more readily, the greater the number of layers of the atmosphere of different densities which exist at any given place. So, in northern climates, where irregular and high winds may prevail, strata might be produced in great numbers by meteorologic conditions, and as each layer or stratum has a different density from the one above or below it, it would take its part in the total amount of refraction. Where the distribution is fairly equable as it is in the region of the trade winds, we would expect a greater proportion of the waves of short length to pass the atmosphere to the earth; at about 30° of latitude accurate observations will probably discover locations in which the sun has great actinic power. At the equator, where the great mass of heated air rises to flow toward the poles, while the air from the north and south passes in, we may have what might be termed an atmospheric lens effect, serving to concentrate the rays of the sun, as they pass from the rarer to the denser medium, and thus giving more intense insolation. In this discussion the modifying factors of diffused light, and of that reflected from the earth, are not taken into consideration, obviously these will vary greatly, and will be much influenced by local conditions.

Although we may conclude, from the experimental data given in the preceding portions of this paper, that insolation in our regions is really of greater intensity than it is in northern climates, we must also note that the measurements of value in determining this question are undertaken on days of bright sunlight or of slight haze. It has already been shown that the latter are often more "actinic" than the former, perhaps because of reflection of the sun's rays which reach the earth, thus increasing the total quantity of light over that which would be present on a clear day. This, however, does not account for the differences observed between two equally clear days at the same time of the year, one of which may be "actinic," the other "nonactinic."

What causes this difference? Is it brought about by air currents of differing density, causing refraction or even total reflection of a portion of the light? Is it due to some disturbance in the chromo- or photosphere of the sun itself? Or, again, has the phenomenon of ionization of the air, by absorbing the radiant energy for this purpose, a connection with these conditions? I confess I am neither physicist, astronomer nor meteorologist, and must leave the answer to others. It suffices here to state that this is a fact which must be reckoned with in discussing the total annual insolation of our regions.

Again, when we consider many places in the Tropics, which are regarded, as a whole, as having a climate unsuited for continued residence of European races, we must remember the large proportion of the year in which the sky is overcast. Cloudy days, in distinction from those in which the sun is but partly obscured by haze, naturally show the effects of insolation to a much less degree than days of sunlight, the intensity

of the light diminishing with the density of the clouds. The Philippine Weather Bureau has stated to me that, so far as they are aware, twenty-four hours never pass on the Island of Luzon entirely free from cloud, and if we calculate the average number of hours of sunshine in Manila as compared with those of many other places in northern climates, such as Denver or Santa Fe, situated in the Middle West of the United States, we find the figures very must less for this district, namely, for Manila, 51 per cent of the theoretical sunshine as against 69 per cent for Denver and 76 per cent for Santa Fe. Even Chicago and New York present figures higher than Manila, namely, 57 per cent for Chicago and 56 per cent for New York. While, during the clear days, the sunlight may be more intense here, nevertheless many northern climates have, on the average, more hours of insolation, entirely regardless of the lengthening of the days during the summer months as we leave the equator. If we take an entire year we may not have a greater amount of radiant energy here than in many places of the Temperate Zone, and perhaps even less.

To what, then, are the supposed untoward effects of the tropical climate due? Is it perhaps that at certain times of the year we may have a greater intensity of light, although the average may not be any more? If this were true, would not the total result be even more apparent in such regions as New Mexico, or in deserts, in which certainly the sum total of insolation would be larger in quantity than in Manila? Undoubtedly, periods of great light intensity would, during certain times, have their effect in the more rapid destruction of lower organisms, and on plant life, on which only a brief period of excessive insolation would be either destructive or highly detrimental, but is this true of a highly organized being such as man, who, for a large part of the year, is protected, to a greater or less extent, by meteorologic conditions? The results of intense insolation do not seem to be so very apparent in this region; certainly cases of sunstroke are rare.

Much has been done in the way of experiment with clothing and head coverings of various colors, so as to avoid these very same effects, but in introducing any such modification the experimental work of Baly and his co-workers[22] in regard to the absorption spectra for ultra-violet of the various dyes must be taken into account. It does not matter so much what the color of the dye is, so long as it presents a considerable absorption band in the proper region of the spectrum.

It seems to me that in any discussion of the effects of the Tropics, not only the sunlight, but also the general average humidity, the proportion of cloudy and rainy days, and above all, the continued equable temperature without sharp contrasts of heat and cold, must be taken into

[22] *Journ. Chem. Soc. London* (1904), 85, 1029; (1905), 87, 1332; (1906), 89, 502, 966, 982; (1907), 92, 1572.

consideration. Then, too, we must remember that the hygienic surroundings are different here than in our home countries, and in the majority of districts lying in these regions the native population has not, as yet, advanced sufficiently in education to eliminate many of the factors which in themselves may produce a marked effect. Perhaps, when modern methods have been more generally extended, the world may to a large extent alter its opinion in regard to this portion of the globe. Europeans, coming to the Tropics, change their mode of life, and do many imprudent things which bring their after-effects.

The subject which I have undertaken to discuss is so complex, the experimental work is as yet so little advanced, that I have been able to give only an outline of what may be done, and to suggest lines of investigation for the future. Many of the papers or monographs which have a more or less direct bearing on it I have not been able to quote at all for lack of space. The physicist, chemist, meteorologist and experimental biologist should all combine to bring a clearer understanding into the field; the plant physiologist and ecologist certainly have topics for study for many years to come. If my paper may seem to be inconclusive and simply to bring together a number of nonrelated results, it is because the subject can not, at present, be treated in any other way.

Work on the sunlight will be continued in the Bureau of Science as opportunity affords, and we hope, in the not too distant future, to bring greater clarity into the field. The biological laboratory, with the means now at hand, will also study the effects of tropical light on microörganisms.

ILLUSTRATIONS.

PLATE I. Spectra of various metals and of the sunlight taken with the grating and quartz lens. The lines in the ultra-violet are obscured by reproduction. The print is given to show, approximately, what the instrument will do, and a more complete series will be published in another communication, when better results are obtained.

TEXT FIGURE.

FIG. 1. A. Lateral view of spectroscope; B. End view of slit and camera; C. Grating; D. Quartz lens; E. Slit.

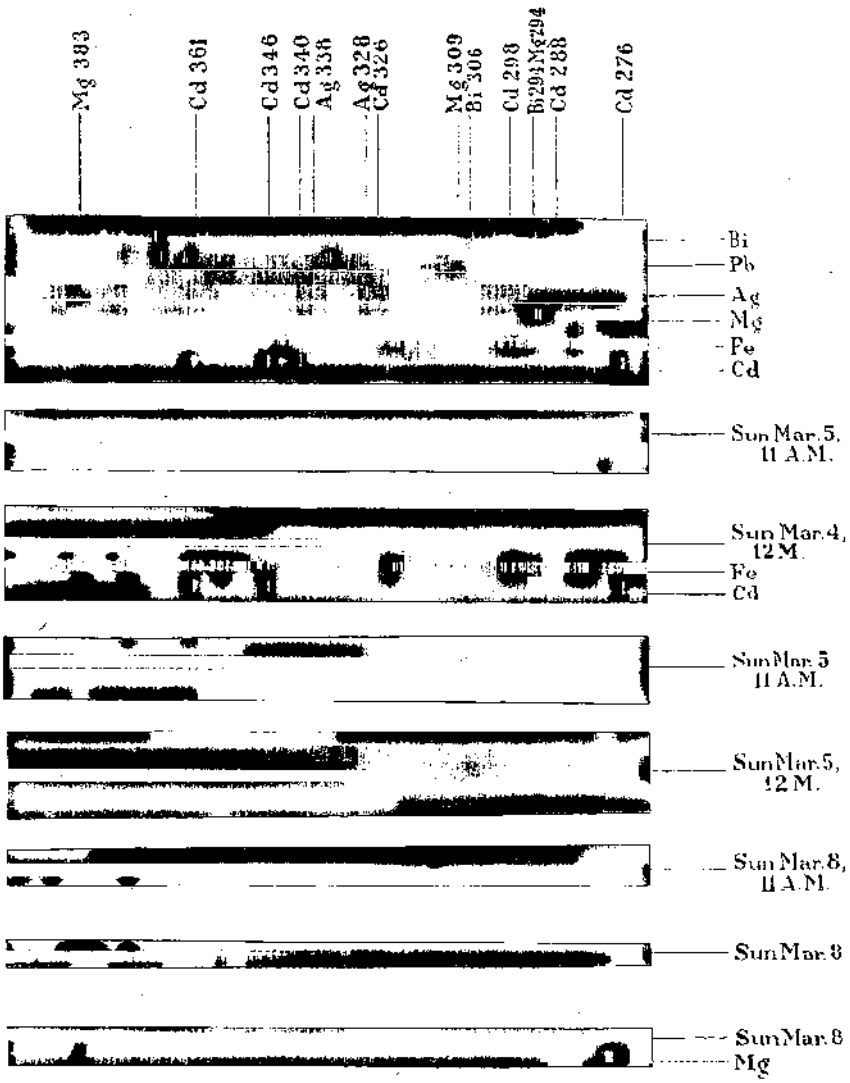
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PLATE I.

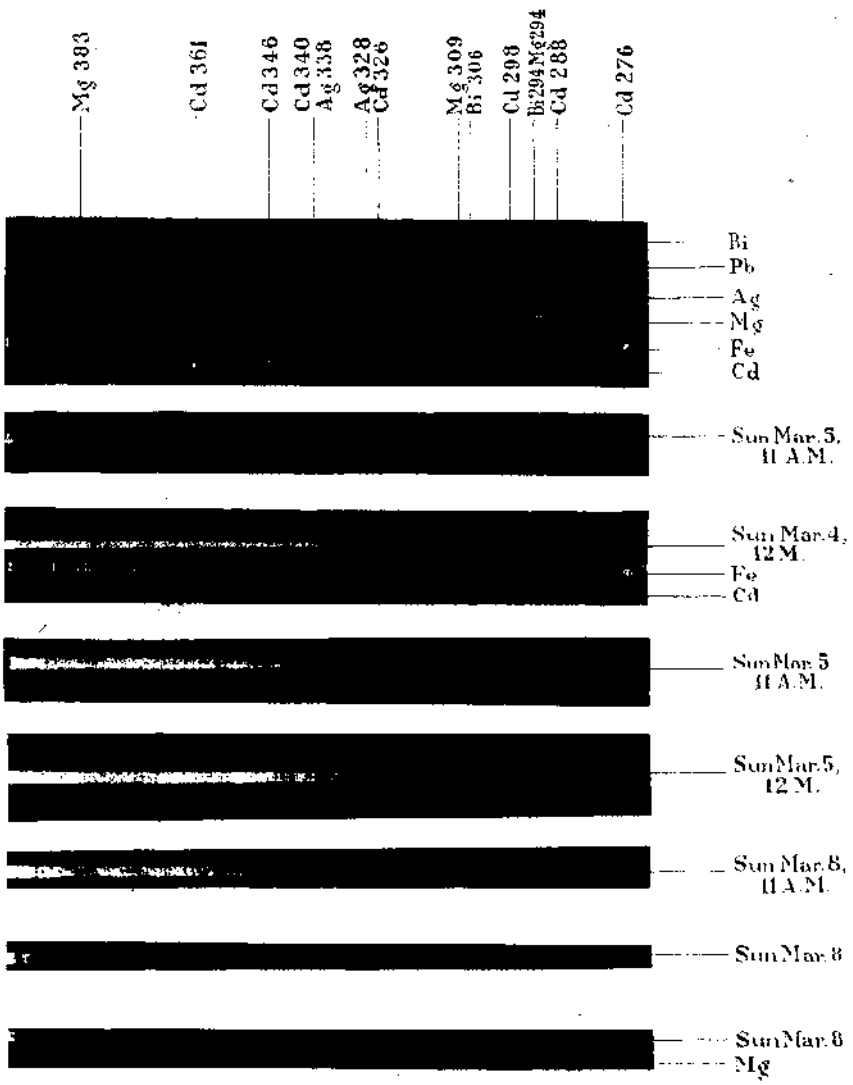
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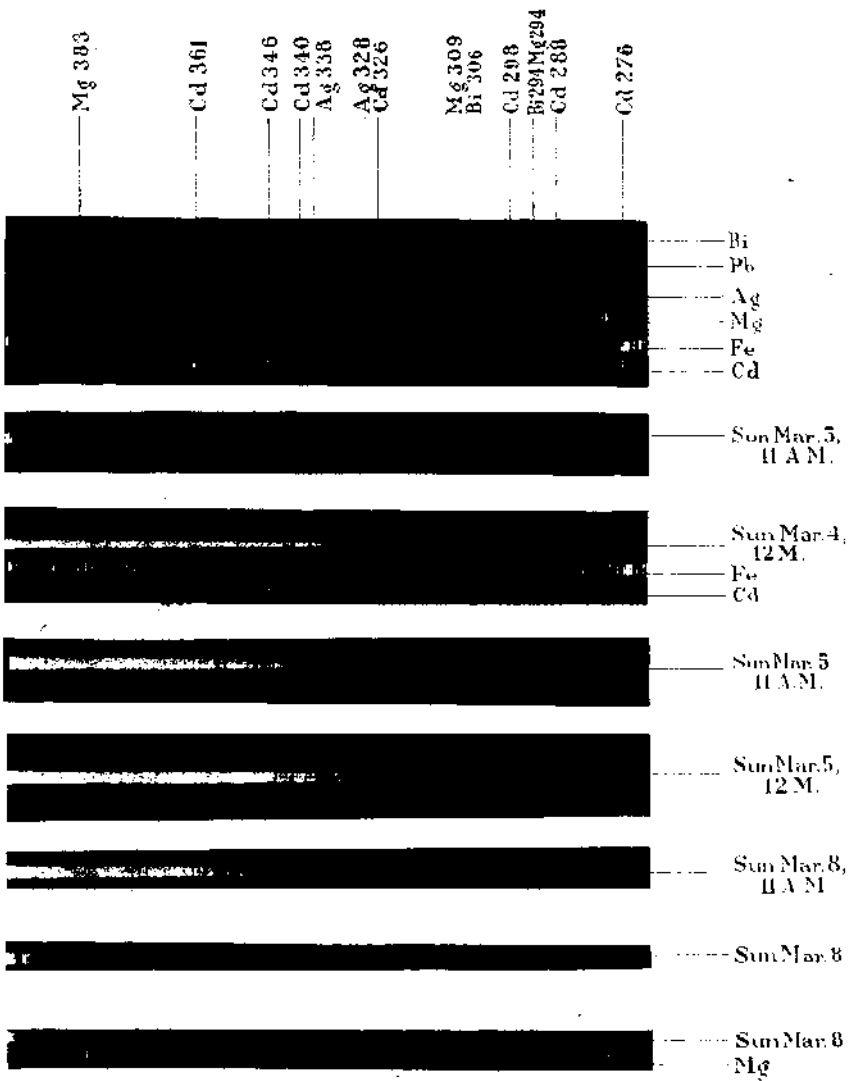
PLATE I.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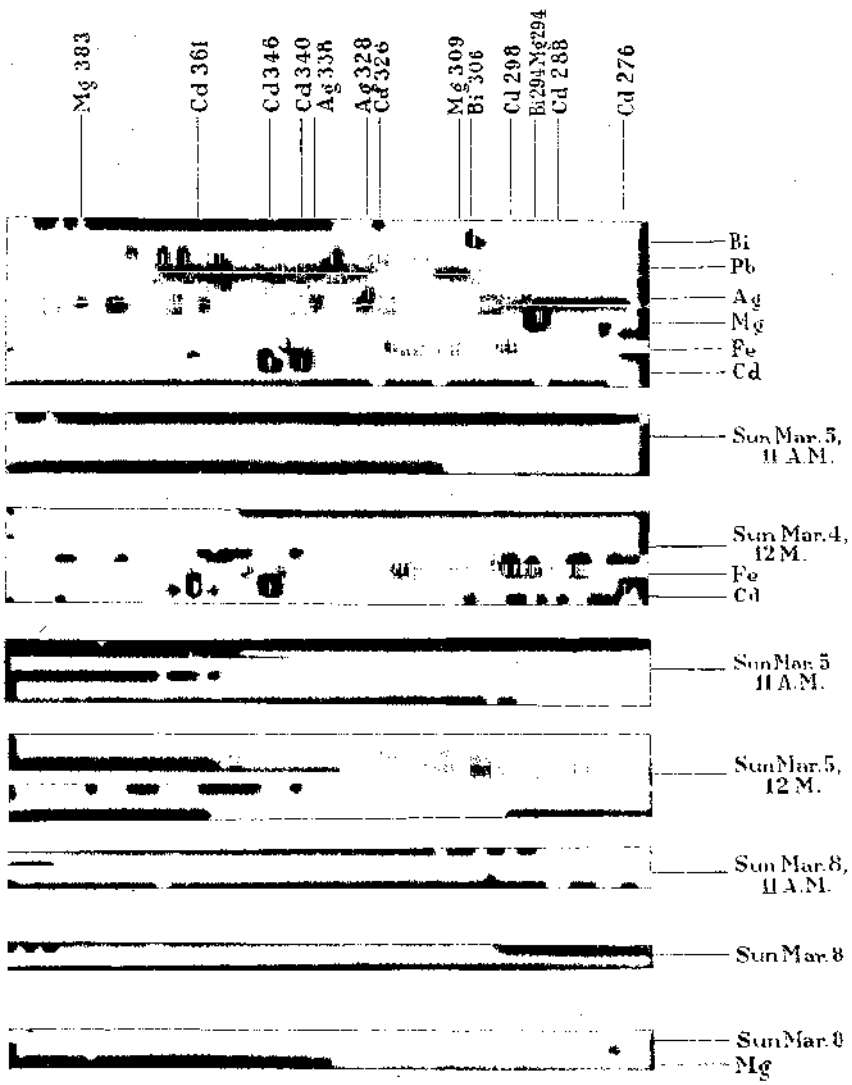
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PLATE I.

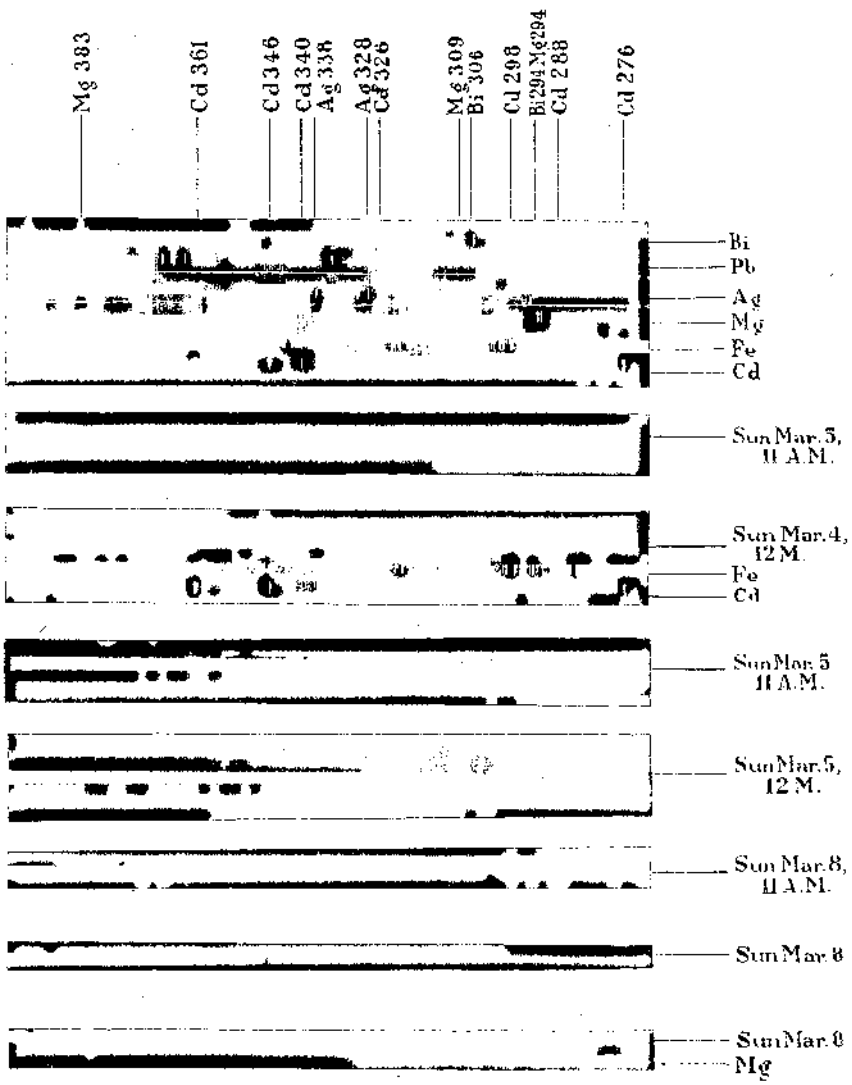
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PLATE I.

THE TREATMENT OF TRYPANOSOMIASIS WITH ESPECIAL REFERENCE TO SURRA.[1]

By Richard P. Strong and Oscar Teague.

(From the Biological Laboratory, Bureau of Science, Manila, P. I.)

One of the most important problems that confronts the scientific medical world to-day is the treatment of trypanosomiasis. Sleeping sickness is claiming its thousands of victims each year in Africa. New districts are becoming infected and the disease is on the increase in certain others in spite of the precautions that are being taken. From an economic point of view the loss of horses, cattle and camels from surra and other forms of trypanosomiasis is very large. India, Egypt, Java and the Philippines have suffered severely in this respect. The losses during the epidemic which began in Manila in 1901 amounted to several millions of dollars. Almost all of the large islands of the Philippines have been shown to be infected from time to time with surra, and the loss in horses and cattle has been constant and of no small magnitude. From a theoretical point of view, studies in the treatment of trypanosomiasis have led to the development of a new field in therapeutics, viz, that of "chemotherapy." Since the trypanosomata are motile and live for several hours in defibrinated blood, it is possible to study the action of various chemical substances upon them; such of these substances as prove to be effective against the parasites *in vitro* may then be tested by injection into infected laboratory animals. By these methods it can readily be determined whether or not changes of a given nature in the constitution of an organic compound improve its efficacy against the trypanosomata.

In the following discussion we shall not consider the development of the therapy of trypanosomiasis in chronological order, nor attempt to consider the entire literature upon this subject, but, for the sake of clearness, we shall arrange the more important methods of treatment in the following order and discuss each separately:

I. Serum therapy and vaccination.
II. Treatment with aniline and other dyes.
III. Treatment with compounds of arsenic.
IV. Treatment with compounds of antimony.
V. Treatment with a combination of two or more drugs.

[1] Read at the first biennial meeting of the Far Eastern Association of Tropical Medicine, at Manila, March 7, 1910.

I. SERUM THERAPY AND VACCINATION.

Antibodies against trypanosomata have been demonstrated in the sera of animals suffering from a chronic form of the disease and it has even been shown that such sera when injected simultaneously with the trypanosomata of nagana into rats will sometimes protect the latter from infection. However, all attempts to obtain a therapeutic serum against the trypanosoma infections have thus far failed. Laveran and Mesnil (1902) tested the effect of injecting human serum into nagana-infected rats and mice on the supposition that man is not susceptible to nagana. It is claimed that marked improvement followed the injection of human serum. However, the disease in these animals was not cured.

The vaccine treatment, that is, the injection of blood containing killed trypanosomata, seems likewise to have no influence on the course of the disease.

Martini suggested that the inoculation of cattle and horses in infected districts with an avirulent strain of the trypanosoma in question might protect them against later infection with a more virulent form of the disease. In our opinion this would be a very dangerous experiment; for it seems possible that the avirulent trypanosomata might regain their virulence in the course of time and that the "parasite carriers" for which we were responsible might become a fearful menace to all the stock of the district. In this connection it is interesting to note that a number of observers have shown that animals which have recovered from trypanosomiasis by treatment are not immune on reinoculation. Manteufel also found that a previous combined treatment with virulent trypanosomata and a highly immune specific serum does not as a rule result in the production of an efficient immunity.

TREATMENT BY DRUGS.

In observing the results of any form of treatment, usually the more acute the course of the disease, the more readily can conclusions be formed in regard to the efficacy of any particular drug. This fact should be borne in mind in the selection of the experimental animals for testing the various remedies. The extremely chronic character of sleeping sickness in man explains why such a long time has elapsed before anything like a unanimity of opinion with regard to the efficacy or inefficacy of certain drugs has been achieved.

Since small animals very generally tolerate larger doses of medicine in proportion to their body weights than do larger ones, theoretically it should be easier to destroy parasites in the blood of the former than of the latter. It will be seen later in the discussion that this actually has been found to be the case. It therefore follows, and should be borne in mind in the following discussion of the literature, that results obtained on the smaller animals with a given drug can not also be assumed to hold good for larger ones.

II. TREATMENT WITH ANILINE AND OTHER DYES.

The treatment of trypanosomiasis with the aniline colors was introduced by Ehrlich, who first employed coloring matters belonging to the benzopurpurin group for this purpose. A substance (trypan red) was discovered which was found to possess marked trypanocidal properties. Since this discovery considerable attention has been given by numerous observers to the study of the value of other coal-tar dyes belonging to the benzidine (diazo) group, to the triphenyl methane, and to the afridol colors.

However, very extensive experiments have shown that while the subcutaneous injections of trypan red, trypan blue, brilliant green, malachite green, parafuchsin and a few other dyes, into small animals, such as mice and rats, can sometimes permanently free them from trypanosomata, larger animals can not be cured by these drugs. Koch treated a number of patients suffering from sleeping sickness in Africa with trypan red but obtained such unsatisfactory results that he soon discontinued the use of the drug. The administration of the dyestuffs by mouth to experimentally infected animals has given even less satisfactory results than the subcutaneous injections. It is true that Ehrlich was successful in preventing subsequent infection in mice after feeding them parafuchsin and that this work was confirmed by Browning. However, recently Breinl and Nierenstein were unable to obtain any favorable results by such a method in large animals. Two horses were fed on parafuchsin; one received 15 grams daily by mouth for thirty days and died after having shown toxic symptoms from the parafuchsin; the second horse received 15 grams by mouth for forty-eight days. On inoculation it became infected in the same manner as an untreated animal. In our opinion the aniline dyes thus far experimented with are of practically no value in the treatment of sleeping sickness in man, or of trypanosomiasis in the larger domestic animals. This field of treatment seems at the present time practically to be abandoned.

III. TREATMENT WITH COMPOUNDS OF ARSENIC.

It appears that Livingston and Braid, as long ago as 1858, first suggested the use of arsenic for the treatment of horses infected with the bite of the tsetse fly.

Lingard, in 1893, and Bruce, in 1894, also employed arsenic as a curative and prophylactic agent for surra and nagana respectively.

Laveran and Mesnil, in 1902, performed extensive experiments with several arsenical compounds, obtaining the best results with sodium arsenate. With this drug they were able to cause the parasite to disappear from the blood. While the lives of the animals were considerably prolonged, a permanent cure was not effected. Similar results were obtained in the treatment, by arsenic, of horses afflicted with surra in the Philippines by Curry, Musgrave and one of us in 1902 and 1903. Moore, Chichester, Thomas, and Breinl also performed extensive experiments with arsenic and sodium arsenate.

All of this work demonstrated that while the course of trypanosomiasis could be modified and the lives of many of the animals prolonged, with the largest animals, at least, a cure did not result and the disease eventually relapsed. Very severe toxic symptoms and necrosis were frequently produced by the drug. Investigators therefore naturally sought for a less toxic preparation of arsenic and for one which was less likely to cause necrosis.

F. Blumenthal had shown that atoxyl when given to rabbits was forty-five times less toxic than Fowler's solution. Several other observers confirmed these results as to the diminished toxicity of this preparation when compared with arsenious acid. Thomas and Breinl first advanced superior claims for atoxyl as a curative agent in cases of trypanosomiasis after a most searching test on animals. They concluded that it was the only remedy at that time known which gave any promise of a cure.

Almost immediately, numerous investigators undertook the study of the value of this drug and the first reports regarding its efficacy in trypanosomiasis were usually very favorable.

R. Koch treated 986 cases of sleeping sickness; out of 356 cases *positive results* were obtained in 347. The cases were divided into early and advanced. Only the former were affected favorably by the drug.

Favorable results were also obtained by Manson in 5 cases of this disease. Kopke did not secure good results. Out of 29 treated cases of sleeping sickness, 22 died. Only 2 were in good health at the time the report was made.

Ehrlich then showed that many of the relapses after treatment with atoxyl were due to the fact that the parasites had become more resistant to the drug named; they had grown to be "arsenic or atoxyl fast." Ehrlich found after the use of parafuchsin, trypan red and trypan blue that resistant strains were also developed. Moore's detailed and careful studies in the employment of atoxyl soon demonstrated that the efficacy of this drug in the treatment of trypanosomiasis was not nearly so great as might have been supposed from the results of the experiments reported earlier.

In 1909 Moore, Nierenstein and Todd, after extensive experiments with atoxyl on dogs, guinea pigs, mice and donkeys, found this drug entirely unsatisfactory and incapable of saving the animal when employed alone. Brienl also concluded that prolonged experience in the treatment of sleeping sickness in man and, to a certain extent in the experimental animals, has proved beyond doubt that atoxyl by itself effects a really permanent cure in comparatively few and only in exceptional cases of sickness in man, even after it has been administered over a prolonged period, and that nearly all the experiments with horses and cattle infected with *Trypanosoma gambiense* lead necessarily to the conclusion that atoxyl alone is insufficient for a successful issue of the treatment and that it only prolongs life to a certain extent, the animals nearly always eventually succumbing to the disease.

Hodges also concluded that the treatment of trypanosomiasis, or of sleeping sickness, with atoxyl is far from satisfactory and states that its use alone has practically been discontinued in Uganda.

More recent experiments in the treatment of trypanosomiasis have been performed with certain derivatives of atoxyl; one of the most important of these is acetylated atoxyl.

Acetylated atoxyl (arsacetin) was first prepared by Ehrlich and Bertheim, who showed that it was much less toxic than atoxyl for several species of animals and that it could be sterilized without decomposition. Browning believed that animals could withstand ten times the amount of arsacetin as of atoxyl. He also believed that the drug was much more effective than atoxyl in the treatment of infected mice. The experiments by Breinl, however, seem to show that arsacetin is less toxic for animals very susceptible to atoxyl, such as dogs, and that proportionately it is not less toxic for horses and guinea pigs. Uhlenhuth and Woithe confirmed the fact that arsacetin was less toxic for rats than atoxyl. Browning and Wendelstadt, who employed the drug in the treatment of rats, noticed marked tremor a long time after the injection of the drug, in the animals which recovered. Wendelstadt concluded that while rats could be cured with arsacetin, in the large doses required to produce a good result, unfavorable symptoms usually appeared. Moore, Nierenstein and Todd found arsacetin superior to atoxyl. However, when used alone, the animals were not cured by this drug.

The only difference from a chemical standpoint between arsacetin and atoxyl consists in the partial acetylation of the amino group. Kopke has reported it to be very probable that in the treatment of human trypanosomiasis one can obtain with arsacetin the same therapeutic result as with atoxyl, perhaps with less risk of poisoning. Eckard has employed the drug in 134 cases of sleeping sickness. Eighty-six of these were improved, but the cases had not been observed for a sufficient length of time for the author to determine definitely the efficacy of the drug. He considered the drug to be as valuable in the treatment of sleeping sickness as atoxyl.

Apparently, no reports of the use of this drug in the treatment of the larger domestic animals have been published. The consensus of opinion seems to be that it possesses about the same, or a little better therapeutic value than atoxyl.

Experiments with other derivatives of atoxyl, such as salicyl atoxyl, formyl atoxyl, sodium *p*-hydroxyphenyl arsenate, disodium azobenzene 4-arsenate, disodium 4-oxyazobenzene 4-arsenate, tetrasodium phenazine 4-arsenate, sodium di-*p*-acetyl aminophenylarsenate (Breinl and Nierenstein) and with substances closely allied, but having instead of an aniline a toluidin nucleus, for example, orsudan and its derivatives, have also been undertaken, but with no more favorable results.

This concludes the list of compounds of arsenic which have already been extensively employed. The use of arsenophenylglycin, a new preparation, will be considered when our experiments are discussed.

IV. TREATMENT WITH COMPOUNDS OF ANTIMONY.

Plimmer and Thomson suggested the use of antimony, an element chemically closely allied to arsenic, in the treatment of experimental trypanosomiasis in 1907.

They employed the sodium potassium and lithium salts of antimonyl tartrate, of which the sodium antimonyl tartrate proved to be the most efficacious. It quickly caused the disappearance of the parasites from the blood after injection.

Of 39 rats injected and treated with the drug, the majority were living fifty-two days later. This interval is obviously too short to enable a judgment

in regard to the final result of the experiments to be made. In more recent work, performed during the past year, these authors state that antimony is as valuable as arsenic in the treatment of trypanosomiasis.

Mesnil and Brimont experimented with potassium antimonyl tartrate in rats and found that the drug caused the disappearance of the parasites from the circulation in about two hours after the injection, but that in many cases they reappeared in the blood. The animals infected with some, but not all, of the species of trypanosomata were cured by this drug.

Boyce and Breinl, however, did not find sodium antimonyl tartrate to give good results either in infected rats or horses. One horse and one monkey were treated with this drug, both eventually succumbed to the disease.

Mesnil and Brimont found in further experiments on mice, that when the animals were treated with tartar emetic, relapses usually occurred, although in the case of the trypanosomata of surra and dourine the parasites disappeared for a time after a single injection of the antimony compound.

Laveran also found tartar emetic unsatisfactory, and sulphide of antimony less active than sulphide of arsenic (orpiment).

Uhlenhuth and Woithe treated 27 rats with sodium antimonyl tartrate. Their results were very discouraging. Repeated injections did not even cause temporary disappearance of the parasites.

Manson has reported the unsatisfactory treatment of one case of sleeping sickness with this drug.

Broden and Rodhain have used soluble as well as insoluble compounds of antimony in cases of sleeping sickness. The hypodermic injections were followed by great irritation and pain so that the drug was given intravenously. Seven hundredths of a gram sometimes caused the disappearance of the parasites from the blood, but they frequently reappeared after a short time. After repeated injections the patients usually lost appetite and complained of malaise. They regard the antimony compounds as about of equal value with atoxyl.

Hodges states that the treatment both with antimony cream and with tartar emetic, singly, has been unsuccessful; that these drugs do not seem to be effective for any length of time and that the treatment of patients with antimony compounds had been discontinued in Uganda at the time the report was made.

Breinl and Nierenstein attempted to prepare an organic antimony compound analogous to atoxyl. After many trials they succeeded in making para, meta and ortho aminophenyl stibinic acids. The ortho compound was impracticable and the meta was unsatisfactory. Extensive experiments with the para compound showed it to be a fairly powerful trypanocide, although not so rapid in its action as sodium antimonyl tartrate. It was less likely to cause abscesses on injection. The authors advise a careful, systematic examination of the urine in the cases treated because of the danger of the production of kidney lesions.

Kopke has treated a few cases of sleeping sickness resistant to atoxyl, with this drug. The injections seemed to cause the disappearance of the parasites, although the author states the inoculations were far too painful for general use.

Laveran has recently suggested the use of an aniline compound of antimony. In potassium antimonyl tartrate potassium was replaced by the aniline radical. Several experiments on guinea pigs seemed to show that the drug was superior to the potassium salt of antimony. A few injections had been made in natives of Senegal, but no final results have been reported.

In the further treatment of sleeping sickness, neither Martin and Rigenbach nor Broden and Rodhain have found tartar emetic administered alone of any great value; the action of the drug after injection is marked, but, as a rule, is only temporary.

V. TREATMENT WITH A COMBINATION OF DRUGS.

A number of investigators who have failed to secure good results in the treatment of trypanosomiasis by single drugs have sought by a combination of two or more to obtain better ones. Some encouragement was given to this idea from the fact that in treating animals infected with trypanosomata, which, for example, had been injected with certain of the dyes or with arsenic compounds and in which dye fast or arsenic fast strains of trypanosomata had developed, injections of antimony seemed more successful owing to the fact that the parasites were not antimony fast. Also, certain drugs which by themselves were inactive against trypanosomata, for example, compounds of mercury or picric acid, when used in combination were supposed to be effective. A large number of experiments have been performed with these various combinations of drugs, but usually with little more definite success than with the remedies employed singly. The following may be mentioned among the combinations of drugs which have been largely employed: (1) Atoxyl and various dyes; (2) atoxyl and bichloride of mercury; (3) atoxyl and tartar emetic; (4) atoxyl and orpiment.

TREATMENT WITH ATOXYL AND ARSENIC SULPHIDE (ORPIMENT.)

Laveran and Thiroux found that a combination of trisulphide of arsenic or precipitated orpiment with atoxyl gave encouraging results in the treatment of guinea pigs infected with trypanosomiasis.

Thiroux and Teppaz treated four horses in this manner, apparently successfully, as all were cured and under observation for six months. The authors state that the animals were infected with a species of trypanosoma other than the one causing surra. Two of the horses were treated with the sulphide alone, neither of these being infected with the surra parasite. Both recovered.

Holmes from India has also reported favorable results with the combined atoxyl and orpiment treatment. Seven guinea pigs and two rabbits infected with trypanosomata received atoxyl and orpiment. In no instance did the trypanosomata reappear in the blood after the first injection of atoxyl. However, four of the guinea pigs died during the period of observation—three months. In the three which lived, four, six and seven doses of the drug, respectively, were given. Seven ponies were also treated with atoxyl and orpiment, given alternately with an interval of one day between each dose. In four, the author reports the treatment to have been successful. Two of the animals of this series and four other ponies not included in this series died of the treatment. One relapsed and was treated with atoxyl, orpiment and tartar emetic for fifteen days and was stated to be cured. Six other ponies were treated with a combination of atoxyl and tartar emetic plus orpiment and sodium arsenate. In three of this series the results were not successful. A number of animals showed relapses before they were cured. The doses of orpiment employed were much smaller than those recommended by Laveran and Thiroux, who administered up to 30 grams. The maximum dose employed by Holmes was 3 grams of commercial arsenious sulphide of a light yellow color. S. H. Gaiger has reported on the treatment of camel surra with red and yellow orpiment. The results were entirely unsatisfactory. In one camel treated with atoxyl and orpiment the result also was not successful.

TREATMENT WITH TRYPAN RED AND ARSENIC.

In 1905, Laveran first suggested a combination of trypan red and arsenious acid in the treatment of trypanosomiasis. He had obtained good results in mice, rats and monkeys. Franke also strongly recommended the use of trypan red and arsenic for this treatment.

Wendelstadt and Fellner suggested a combination of arsenic and brilliant green and brilliant green and nucleic acid. Magalhaes did not obtain favorable results with sodium arsenate and brilliant green.

Thomas gave up the combined arsenic and dye treatment because of the nephritis and local necrosis so frequently caused by it, although Breinl and he thought that a combination of arsenic and an improved form of trypan red, if it could be obtained, would seem indicated in the further search for a cure of trypanosomiasis.

TREATMENT WITH ATOXYL AND MERCURY.

Moore, Nierenstein and Todd obtained the best results with injections of bichloride of mercury after the parasites had been caused to disappear from the peripheral circulation by treatment with atoxyl. While in small animals the results were favorable, in larger ones the drugs were not sufficiently efficacious to be of practical value.

Plimmer and Thomson found that the combination of atoxyl and succinimide of mercury was most successful. In a later communication, however, their results were not so favorable. Where sufficient doses of the compound of mercury were given, chronic lesions of the kidney and liver were observed.

Moore, Nierenstein and Todd in further experiments with rabbits and donkeys infected with *Trypanosoma gabiense*, used atoxyl and bichloride of mercury. The outcome of this work was, to use the authors' own words, extremely disappointing. It was found impossible to save a single animal. Of 5 rabbits infected with *Trypanosoma brucei* and treated in the same way, 4 were apparently cured. These authors also performed experiments on dogs, guinea pigs and mice with arsacetin (acetylated atoxyl), followed by bichloride of mercury. They considered treatment with arsacetin followed by a compound of mercury more efficacious than treatment by arsacetin alone, and arsacetin of more value than atoxyl, but believed none of these methods to be of practical value since death invariably occurred.

These authors also employed combinations of atoxyl-silver nitrate; atoxyl-lead acetate; atoxyl-quinine cacodylate; atoxyl-potassium bichromate; atoxyl-quinine.

These were found to be valueless. Treatment of mice infected with *Trypanosoma brucei*, with trypan red followed by bichloride of mercury, was superior to trypan red alone, but inferior to the combined treatment with atoxyl and bichloride of mercury.

Laveran and Thiroux also used a combined treatment with atoxyl and bichloride of mercury on guinea pigs infected with the surra parasite. Their results were not particularly favorable, although better than with the treatment by atoxyl alone.

Uhlenhuth, Hübner and Woithe report that in rats the combined treatment was of considerable value.

Gray described a thorough trial of the treatment of sleeping sickness with atoxyl and bichloride of mercury. The results obtained were much superior to those with atoxyl alone, but were far from satisfactory.

Broden and Rodhain found that the combined treatment with atoxyl and bichloride of mercury did not prove itself at all superior to that with atoxyl alone.

Breinl employed acetylated atoxyl, sublimate and Donovan's solution, in 6 monkeys. Five out of the 6 animals were alive and free from parasites at the time the report was made.

Hodges states that the results of the treatment of sleeping sickness with atoxyl and bichloride of mercury do not, at present, promise to be of a more permanent nature than do those with atoxyl alone.

TREATMENT WITH ATOXYL AND ANTIMONY.

Broden and Rodhain, and Martin and Darné, in the treatment of sleeping sickness, combined atoxyl with injections of the soluble antimony compounds with very encouraging results in early cases of the disease.

Hodges states that in man, treatment by atoxyl and antimony has not at present shown favorable results in Uganda, although these remedies in combination and alternation are being more extensively tried.

Rennes treated one horse infected with dourine with atoxyl combined with antimony. Two months afterwards the blood of the animal was free from parasites. Six months after the beginning of the treatment, the horse was apparently well. He was then reinoculated and contracted a new infection.

Martin, Leboeuf and Rigenbach believe that atoxyl given with tartar emetic is more poisonous than when given alone, but that the combination is the most effective treatment for trypanosomiasis they have tried. The antimony compound was given intravenously in doses not greater than 10 centigrams of a 1 per cent solution. They treated 31 cases of sleeping sickness with the combination of atoxyl and the antimony compound. Only one of these was in the first stage of the disease. Of the 31, at the time of the report, 10 were dead, 11 had abandoned treatment and 10 were still under observation; 7 of these had parasites in their blood. The authors finally conclude that while the combination of atoxyl and tartar emetic is capable of giving excellent results in early cases, it does not succeed in those which have entered the second stage of the disease.

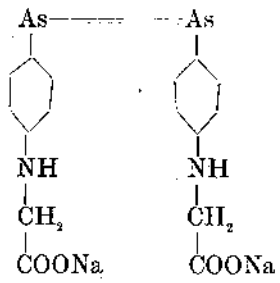
TREATMENT WITH ARSENOPHENYLGLYCIN.

Through the courtesy of Prof. Paul Ehrlich, of Frankfort, who very kindly offered to send us new preparations made in his laboratory and which seemed of most value in the treatment of trypanosomiasis, for further experimentation in the treatment of larger animals and for a demonstration of their practical efficiency, a shipment of arsacetin was first received and, in the beginning of the year 1909, a shipment of arsenophenylglycin. Professor Ehrlich has continued to supply us with repeated shipments of arsenophenylglycin and all of our experiments with this drug have been carried on with the compound prepared and sent from his laboratory. We take this opportunity of expressing our gratitude to Professor Ehrlich for having given us the opportunity to study carefully the effect of this preparation and for so generously supplying us with the drug.

Arsenophenylglycin is a light yellow powder, very soluble in water

and containing about 38 per cent of metallic arsenic (atoxyl contains about 31 per cent).

It has the following formula:



It is considerably less toxic than atoxyl.

The drug was received in sealed glass vacuum tubes. Since it is readily oxidized to a much more toxic, brown substance when exposed to the air, it should be dissolved and used immediately after such exposure. The most favorable strength for injection is from 5 to 10 per cent. It may be given either subcutaneously or intravenously. In man, subcutaneous injection at times calls forth a marked local reaction with subsequent abscess formation. This local reaction is sometimes dependent, according to Ehrlich, upon too strong an alkalinity of the preparation.

Although we now have been experimenting with arsenophenylglycin in the treatment of trypanosomiasis for nearly a year, we have made no previous publication of our results, because we wished to wait until the work had been carried on in sufficient detail and for a sufficient length of time to enable us to draw definite conclusions. A study of the literature of the treatment of trypanosomiasis reveals the fact that many of the publications on this subject have been premature. Remarkable claims have been made in turn for each method of treatment, which have not been borne out by more detailed and extensive experiments carried on for longer periods of time.

Our work with this drug has been confined entirely to monkeys, horses and cattle. In our opinion nothing would be gained by experimenting with smaller animals, since several other methods of treatment had already given satisfactory results in mice and rats, but these had failed to cure the larger animals.

It will be seen from our results with monkeys and horses that arsenophenylglycin is the most nearly ideal drug in the treatment of trypanosomiasis that we as yet possess. While our work with this preparation was in progress, several publications on the value of this drug in the treatment of experimental trypanosomiasis in animals have appeared and these will now be reviewed briefly.

Wendelstadt and Roehl have both shown that a single injection of arsenophenylglycin is capable of curing severe trypanosoma infection in mice and rabbits. Roehl has also demonstrated the prophylactic effect of the drug in these animals.

Schilling and Jaffé also have shown the unquestionable value of arsenophenylglycin in the treatment and cure of mice and rats infected with the nagana trypanosoma, the animals frequently being cured by a single injection of the drug. In their experiments with horses, all of the animals died, apparently from the effect of the drug in the doses employed.

Mesnil and Kerandel inoculated three monkeys infected with *Trypanosoma gabiense* with arsenophenylglycin. In two, the parasites disappeared and had not returned after about three and one-half months. In the third a relapse occurred, then the second injection was given and for the following two and one-half months the animal was free from parasites. They also demonstrated the prophylactic value of the drug in several monkeys and have obtained good results with it in the treatment of four guinea pigs infected with the trypanosoma of Togo.

Eckard has treated 19 cases of sleeping sickness with arsenophenylglycin. Twenty-four hours after the injection, the parasites could no longer be found in the patients' blood. The author was unable to state definitely the value of the drug, owing to the short time which had elapsed between the time the injections were made and the date of the report.

Breinl and Nierenstein report experiments with 20 rats, each of which received the dose of arsenophenylglycin recommended by Roehl, 0.4 gram per kilo or a smaller dose. All died from the effect of the drug. Of 5 guinea pigs infected with trypanosomata and treated, 3 died; 2 from the effects of the drug, and 2 were alive and well after 240 days. Three monkeys were treated with a single dose. Two were alive and well 270 days later. One died from the effects of the drug. Dogs could also be cured of infection by repeated injections, even in advanced cases. Four donkeys and 1 pony were treated with subcutaneous inoculations of the drug. One donkey died after a dose of 0.1 gram per kilo. Another succumbed apparently to the local gangrene caused by the drug. In the other three, large injections did not save the animals which died later of infection. These authors believe that the drug is superior to atoxyl, but that the chance of successful treatment with it diminishes as the size of the animal increases.

Zupitza has employed arsenophenylglycin in 25 cases of sleeping sickness. The treatment appears to be the most favorable which had been attempted at the time the report was made. The parasites disappeared from the blood more quickly after the injection of arsenophenylglycin than after any other drug employed. Relapses had occurred in some of the cases. The time of observation had been too short to draw any more definite conclusions. Twelve horses and 7 donkeys were also treated with this drug. The statement was made that one-half of them had been at work for 5 months after the discovery and treatment of the infection. At present Zupitza's article is not at hand and we have only been able to consult a review. We can not therefore conclude whether these animals were free from trypanosomata and had been cured.

EXPERIMENTS WITH ARSENOPHENYLGLYCIN IN MONKEYS.

Dosage.—One of the first questions which seemed necessary to answer was the dosage of arsenophenylglycin most favorable for the treatment of these animals. Ehrlich had suggested to us about 0.1 gram per kilo of body weight. However, when this amount was injected into normal monkeys, it was found to kill far too high a percentage of the animals for the dose to be practical for treatment. Hence we found it necessary to employ the drug in smaller quantities.

In our preliminary experiments with this preparation, 53 monkeys were employed. Our results were very unsatisfactory, for our animals very frequently died, either from the toxic effects of the drug, or from trypanosomiasis, due to the fact that sufficiently large doses of arsenophenylglycin to cure the infection had not been administered. Later we learned that this first lot which we received was an inferior product in comparison with the drug as prepared by Ehrlich at a later date. For this reason the detailed experiments and the protocols of these 53 monkeys are omitted in this paper. It is believed that their introduction would only cause confusion, as the results obtained are now of little value as compared with those derived from our later and most complete experiments with arsenophenylglycin. Mention here of these preliminary experiments is merely made in case that other observers have encountered similar results with an inferior sample of the drug. A short time after these first experiments were carried out, a second shipment of arsenophenylglycin was received and the fatal dose for monkeys accurately determined with this preparation.

The monkeys employed in these experiments, *Cynomolgus philippinensis* Geoff., were in good condition, having been recently captured. The results in determining the dosage are given in Table I, from which it may be seen that the minimum lethal dose equals 0.22 to 0.26 gram per kilo of body weight of animal. The drug was administered in 5 per cent solution, subcutaneously. No local reaction of any consequence was noted following our injections into monkeys. In using the arsenophenylglycin, the sealed tubes were opened, the drug dissolved at once in distilled water and immediately injected. Those animals which died with acute symptoms from an overdose of the drug usually lived only for from two to ten days after the injection. Some of them suffered with bloody diarrhœa before death. At autopsy, the liver and kidneys usually showed marked fatty degeneration, the lesions present being similar to those which have been regularly described in poisoning from atoxyl.

Treatment.—After determining the most favorable dosage necessary in monkeys, we proceeded to treat animals which had previously been infected with the trypanosoma of surra. In every instance the parasites were present in the blood of the animal at the time the treatment was undertaken.

Discussion of Table II.—Table II gives the results observed with four series of monkeys, the individuals of a given series having been inoculated with the same surra blood at the same time and treated at the same interval after the infection.

SERIES I contains 20 monkeys, 10 of which had been recently captured, the others having been in captivity in the laboratory for variable periods of time. Many of the latter had been used for other experiments and were not in good condition at the time of the present inoculation. For

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this reason both sickly and robust monkeys were arranged in each group, receiving the same dose of the drug. The strain of trypanosoma employed for inoculating the monkeys of this series was a virulent one, originally obtained from a native pony which showed well-marked symptoms of surra; it had been passed successively through 2 horses, 1 monkey, and then through a third horse before being employed in these experiments. No attempt was made to treat any of these "passage" animals until after the strain of surra had been passed to the next host, and hence the trypanosomata, when inoculated into the monkeys of this series, had never been in contact with arsenic.

About 3 cubic centimeters of the blood of the last "passage" horse, in which the trypanosomata were fairly numerous, were suspended in 100 cubic centimeters of salt solution, and 2 cubic centimeters of the suspension were inoculated subcutaneously into each of the 20 monkeys on July 8, 1909. On July 12, 1909, a blood examination proved to be positive in nearly every instance, and on July 13, when the treatment was begun, trypanosomata were present in the blood of all the monkeys, being very numerous in most instances.

Two of the monkeys of the series were left untreated to serve as controls, and these died, one seventeen and the other fourteen days after inoculation. Monkey No. 4293, which had been in captivity at the laboratory for a long time, was found dead on the third day after inoculation.

The arsenophenylglycin was administered subcutaneously in doses varying from 0.04 to 0.09 gram per kilo of body weight. Monkeys Nos. 4332 and 4341 were animals which had been employed previously for other experiments and were in poor condition at the time of the present experiment; the former died in eight days, the latter in five days after the treatment, death being due, in our opinion, to the combined effect of exhaustion from prolonged captivity and the surra infection and to the toxic effect of the drug. The results obtained with these 2 monkeys should be disregarded.

With these exceptions, it is seen from Table II that every monkey receiving an injection of 0.06 gram per kilo or less showed a relapse of the infection after an interval of from eighteen to fifty-five days. As soon as they were found to have suffered a relapse, all of the monkeys with the exception of No. 4421, were given a second dose of 0.08 gram of arsenophenylglycin per kilo. With the exception of monkey No. 1473, which died within four days after the second injection, all were, we believe, permanently cured. Two are still alive seven months after the second treatment. Many have become somewhat weak and emaciated from their long confinement in small cages. However, the emaciation was not due to a relapse of the surra infection which remained undiscovered, as was shown by the following observation:

Three of the treated monkeys (Nos. 4479, 4476, and 4481) which were so thin and weak that it was obvious they could live under existing conditions only for a few weeks longer, were taken on November 11, 1909, to the residence of one of us and kept in the open air on a chain. They all quickly began to gain in strength. One died a few weeks later after having been exposed to wind and rain. The other two gained in weight and strength and are still alive.

We believe, therefore, that a number of our monkeys which, although cured, finally died after long periods of captivity, could have been kept alive by placing them under better conditions with regard to food and environment and that this should be kept in mind in studying the tables. Monkey No. 4432, although still free from parasites, was losing in weight and strength and, in view of the possible existence of a latent infection with surra, on October 12 a dose of 0.15 gram per kilo of arsenophenylglycin was administered. This monkey was found dead on the morning of October 22, 1909, its blood having been examined on October 21 with negative results. It seems extremely probable that if the third treatment had been omitted and this monkey had been placed under more favorable conditions, it would not have succumbed.

Of the monkeys receiving 0.07, 0.08, and 0.09 gram per kilo, some were permanently cured by the single inoculation, while others suffered a relapse and were treated a second time with a larger dose. Four of these monkeys have remained free from trypanosomata for eight and two-thirds months after a single treatment with the drug.

A summary of the experiments of Series I shows that a single injection of 0.04 to 0.06 gram of arsenophenylglycin per kilo of body weight causes a disappearance of the trypanosomata from the peripheral circulation, but that the parasites reappeared after a variable length of time; that a dose of 0.07 to 0.09 may effect a permanent cure or may not, but that the majority of monkeys so treated recover. Furthermore, not a single relapse occurred after the administration of 0.08 gram per kilo as a second treatment.

SERIES II.—The surra trypanosoma employed in this series was obtained from a native pony that had acquired the infection under natural conditions. An American horse was inoculated subcutaneously with the blood of this pony and six days later his blood was used for the inoculation of the monkeys. On August 4, 1909, 1 cubic centimeter of the horse's blood was suspended in 20 cubic centimeters of salt solution and 2 cubic centimeters of the suspension given subcutaneously to each of the 8 monkeys. The untreated control monkey died of the infection twenty-six days after the inoculation. The other monkeys were treated on August 10, 1909, and all were given a dose of 0.08 gram of arsenophenylglycin per kilo; only one of these (No. 4559) was definitely shown to have suffered a relapse. One animal, No. 4561, received only a single injection of the drug and is still alive, seven months after the treatment. The majority of the other monkeys were given a second treatment with a larger dose (0.1 to 0.15 gram per kilo); one of these is still alive, the others, apparently on account of their weakened condition, succumbed to the toxic effects of the drug.

Monkey No. 4565 was free from trypanosomata November 16 (three and one-half months after treatment); it has since escaped.

The experiments of Series II indicate that the majority of the monkeys treated with a dose of 0.08 gram per kilo will recover from trypanosomiasis, and, hence, the results obtained in Series I are confirmed.

SERIES III.—The strain of surra employed in these investigations was obtained from one of the horses on the Government stock farm at Alabang where an epidemic of surra existed. From this horse the strain was inoculated into a monkey. On September 9, 1909, 1 cubic centimeter of the monkey's blood containing numerous trypanosomata was added to 200 cubic centimeters of salt solution and 1½ cubic centimeters of the suspension injected into each of 21 monkeys.

Two of the untreated monkeys which served as controls died twenty-two and twenty-three days, respectively, after the inoculation with surra blood. A third monkey, No. 4630, was already moribund from the infection at the time when the treatment of the other monkeys was begun, and may therefore be regarded as a third untreated control.

On September 14 trypanosomata were found in the blood of all the monkeys of the series and treatment in doses of 0.1 to 0.26 gram per kilo, of arsenophenylglycin was administered. None of these monkeys have suffered a relapse and none have received a second treatment. Eleven of them are alive and free from trypanosomata, after five and two-thirds months.

One (monkey No. 4616) was free from parasites for four and one-half months and was then lost. Monkey No. 4625 died after three months, its blood being free from parasites just before death. The other animals of the series which died, apparently succumbed from the effects of the drug.

Series III demonstrates clearly that monkeys receiving single doses of from 0.1 to 0.26 gram of arsenophenylglycin are permanently cured of the disease, although some of them will die from the toxic effects of the drug. Doses of from 0.1 to 0.2, inclusive, effect permanent cures with only a small percentage of deaths.

SERIES IV.—On September 22, 1909, the monkeys of Series IV were inoculated with the blood of monkey No. 4632, which constituted a control animal employed in Series III. Trypanosomata were found present in the blood of these animals on September 27 and all but one, which served as a control, were treated on September 28. The untreated control monkey died from the surra infection seventeen days after inoculation.

Very large doses of the drug (0.26 to 0.36) were administered and the three animals which survived these doses have remained free from parasites for six months.

Summarizing these results, it may be stated that not a single monkey which received 0.1 gram of arsenophenylglycin per kilo or a greater amount has shown trypanosomata in the blood twenty-four hours after

treatment was given and at no time subsequently have the parasites reappeared. From Table I it is seen that the minimum lethal dose is from 0.22 to 0.26 gram, which is more than twice the dose shown to be efficacious in producing a cure in monkeys.

These experiments, therefore, show conclusively that monkeys, *Cynomolgus philippinensis* Geoff., can be cured permanently of surra infection by a single injection of arsenophenylglycin.

EXPERIMENTS IN HORSES.

Early in the year 1909 attempts were made to treat horses infected with surra and brought to the laboratory. The animals were placed in a small screened stable where they were not kept under the best hygienic conditions. Here their weight could only be estimated approximately, and hence the dosage of the drug administered could not be properly controlled. Many of the horses died from the toxic effect of the drug; others during the course of the treatment suffered a relapse of the infection and were either killed or succumbed during further treatment. In some instances a relapse occurred after the animal had survived a large initial dose. The repeated administration of small doses of arsenophenylglycin gave very unsatisfactory results. None of these animals survived. Later we were able to obtain the weight of 5 horses before beginning treatment, with these the results were as follows:

Weight of horse.	Dose in grams per kilo.	Remarks.
460 kilos	0.05	Recovered from effects of drug.
580 kilos	.052	Died in 8 days.
521 kilos	.058	Recovered.
430 kilos	.065	Died suddenly 24 days later.
371 kilos	.066	Died in 2 days.

From these preliminary experiments it appears that the minimum lethal dose for a large American horse is about 0.052 gram per kilo. A horse which died after receiving this amount per kilo was a very fat animal which had been foundered, so that the dose was actually relatively higher than the figures indicate.

Our experiments indicate, therefore, that the minimum lethal dose per kilo of body weight is about three times as great for monkeys as it is for horses.

Moore offers the following explanation for the discrepancies in dosage between large and small animals when calculated on the basis of the body weight. He emphasizes the fact that the seat of biochemical activity after the administration of arsenic or antimony is in the intestinal mucosa, and that it would seem probable that this is also the seat of manufacture of the trypanocidal substance. In the larger animals the therapeutic dose is therefore lowered because of the rel-

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atively smaller number of the intestinal cells to take up the drug and the consequent rapid poisoning of the animal if the attempt be made to give the relative dose. The relatively small number of cells must first act upon the drug before it can be turned out as a trypanocide for the parasites.

This hypothesis can hardly be considered tenable without further experimental evidence. According to Moore's theory, the maximum dose is proportional not to the body weight but to the two-thirds power of the body weight.

Following are the details of the earlier experiments carried on with horses:

Horse No. 1.—Native pony. No history as to length of time sick with surra. Head droops and general appearance bad, respirations rapid. Slight œdema of abdomen, mucous membranes of mouth and tongue pale. Examination of blood on April 22 with one-twelfth oil immersion shows about 1 trypanosoma to a field.

April 24, 4.8 grams of arsenophenylglycin dissolved in 100 cubic centimeters of distilled water and injected subcutaneously.

April 25, blood examination for trypanosomata negative.

April 29, second injection of 6 grams of arsenophenylglycin subcutaneously.

May 3, third injection of 6 grams of the same drug intravenously.

May 3, just before the injection of the drug, a monkey was inoculated with 20 cubic centimeters of the blood of the horse. Repeated examination of the blood of the monkey never revealed trypanosomata.

May 12, 10 cubic centimeters of horse's blood were injected subcutaneously into a monkey. This monkey's blood was examined at intervals for a month with negative results. However, the horse grew weaker, and was found dead on the morning of May 18.

Although we were unable to demonstrate any trypanosomata in the blood of this animal, nevertheless, we believe that it succumbed from the effects of surra. This was the first animal treated by us, and the doses administered were evidently entirely too small for a cure to result.

Horse No. 2.—April 23. This animal was injected with 20 cubic centimeters of the blood from horse No. 1 and which contained trypanosomata. The animal died on May 11 of surra, being untreated and used for the purpose of keeping at hand a virulent strain of trypanosoma for the infection of other animals.

Horse No. 3.—Native horse; suffered with advanced symptoms of surra. Marked œdema of the abdomen. Numerous trypanosomata present in the blood.

April 24, 4.8 grams of arsenophenylglycin given subcutaneously.

April 25, blood examination negative for parasites. Parasites did not reappear in the blood.

April 29, 2 monkeys inoculated with the blood of this horse. Trypanosomata did not subsequently develop in the blood of either of them. A second subcutaneous injection of 4.5 grams of arsenophenylglycin was given and on May 3 another injection of 10 grams intravenously. On May 3, 1 monkey and May 7, 2 other monkeys were also inoculated with the blood of the horse. All these animals remained negative for trypanosomata for over two months. The horse died six days later (May 9), notwithstanding the fact that no trypanosomata were found in its blood. Nevertheless, this animal probably died of surra infection and from the toxic effect of the last dose of the drug.

Horse No. 4.—Large native horse; received on April 29. Blood examination positive for trypanosomata. No record of how long the animal has been sick with surra. Temperature 40°. April 29, 8 grams of arsenophenylglycin in-

jected subcutaneously. Considerable œdema at the site of the injection on the following day.

May 3, monkey was inoculated with 30 cubic centimeters of the blood of the horse, but no trypanosomata developed in its blood. Following the inoculation of the monkey 15 grams of the drug were given intravenously to the horse, although the blood examination the previous day had been negative for parasites.

May 12, a second monkey was inoculated in the same manner with the same result.

June 4, a third monkey was inoculated with 20 cubic centimeters of the horse's blood, on the 15th of June it developed trypanosomiasis and died on the 26th.

This experiment demonstrates that this strain of trypanosoma had not lost its virulence, although the animal from which it was taken had received previously 2 doses of arsenophenylglycin.

June 10, an examination of the horse's blood showed a few trypanosomata. On the same day another inoculation of 10 grams of arsenophenylglycin was given intravenously. The parasites disappeared from the blood, but reappeared on July 6 when 11.4 grams of arsenophenylglycin were again given intravenously. Although the trypanosomata did not reappear in the blood, there was given on July 15 another dose of 10 grams of arsenophenylglycin intravenously and on August 3, 11 grams more. Thirty cubic centimeters of the blood of the horse were injected into a monkey on August 3 before the inoculation with the drug. The animal later developed trypanosomiasis. On August 5 the horse was unable to rise and on August 6 he died, evidently succumbing to the surra infection. Although this animal was treated in a screened stable, nevertheless it was necessary to place him alongside another animal which had surra and which remained untreated. It is possible, therefore, that he may have been reinfected with trypanosomiasis from the other surra animal.

Horse No. 5.—Small bay horse; contracted surra naturally. At the time of treatment, April 29, there was œdema of the abdomen and trypanosomata were numerous in the blood. Ten grams of arsenophenylglycin were injected subcutaneously. On the following day there was considerable œdema at the site of the inoculation. On the day following the injection there was watery diarrhœa which lasted about twenty-four hours. Although no trypanosomata reappeared in his blood, he was given a second inoculation of 10 grams intravenously on May 3. Following this, the animal was unable to stand and there was marked tremor of the muscles. The animal died on May 8, evidently of arsenic poisoning. Three monkeys were inoculated with his blood, 1 on May 3 with 20 cubic centimeters, 1 on May 7 with 10 cubic centimeters, and 1 on May 7 with 2 cubic centimeters. None of these animals developed surra infection.

Horse No. 6.—Native pony; contracted surra naturally. The duration of the disease unknown, although the condition of the animal appeared to be good at the time treatment was begun. On April 30 its blood was positive for trypanosomata and 6 grams of arsenophenylglycin were injected subcutaneously, after which the parasites disappeared from the circulation and did not reappear.

On May 4, 15 grams of the drug were injected intravenously. On this date, previous to the injection of the drug, a monkey was also inoculated subcutaneously with 30 cubic centimeters of the horse's blood. This animal did not develop surra. On May 10 there was considerable œdema along the lower margin of the abdomen. The blood remained negative for trypanosomata. The animal died May 28. The immediate cause of death of this animal is not clear.

Horse No. 7. (Treated with arsacetin.)—Native horse which contracted the disease naturally. Condition good at the time treatment was commenced. No information was obtainable regarding the time the animal had been infected with

surra. Blood examination April 30 was positive for trypanosomata. On this date 8 grams of arsacetin were given subcutaneously. The parasites disappeared from the animal's blood and remained absent. On May 5 there was given a second inoculation of 15 grams of arsacetin intravenously. The horse died on the following day of arsenic poisoning. A monkey inoculated on May 6 with 20 cubic centimeters of its blood did not develop surra.

Horse No. 8.—Chinese horse, infected artificially in the laboratory. Inoculated subcutaneously with 10 cubic centimeters of the blood of horse No. 2 just before it died of surra. Horse No. 8 was untreated and died of surra on June 15. On June 4, June 14, and May 17, respectively, monkeys were inoculated with the blood of this horse and all developed surra infection and died in nineteen, and seventeen, and eleven days, respectively, thus demonstrating the virulence of the strain employed in these experiments.

Horse No. 9.—Small native horse; infected naturally with surra. No information regarding the length of time of infection. On May 21 the animal was injected with 10 grams of arsenophenylglycin intravenously. Trypanosomata disappeared from the blood. On June 12 the animal's temperature rose to 39.8° although no parasites could be found in the blood. A monkey was inoculated with 20 cubic centimeters of the blood and on June 22 was found to have trypanosomata in its blood. Because of the rise of temperature on June 12, the horse was given another injection of 10 grams of arsenophenylglycin intravenously. On June 27, the horse was unable to rise, although the blood showed no trypanosomata. On June 28 a monkey was inoculated with 12 cubic centimeters of the horse's blood. This monkey, however, did not develop trypanosomiasis. The horse died on June 30, the temperature being 40°. This animal also probably died of surra infection.

Horse No. 10.—Native horse; contracted the disease naturally. Œdema of the abdomen and scrotum at the time the treatment was begun. No information regarding the length of time since the animal contracted the infection. On May 21 it was given an injection of 10 grams of arsenophenylglycin. Trypanosomata disappeared from the blood. On June 7, œdema of the abdomen and scrotum was still present. A monkey was inoculated with 35 cubic centimeters of the blood of this horse and died on June 18 of surra. On June 10, the trypanosomata reappeared in the horse's blood, when 10 grams of arsenophenylglycin were given intravenously. On June 29 another monkey was inoculated with 22 cubic centimeters of the horse's blood. This monkey died later of surra.

On June 29 the horse was given 10 grams of arsenophenylglycin subcutaneously and on July 14 he was unable to rise. On July 16 the horse was killed. The examination of the blood and organs were negative for trypanosomata. This animal probably also had surra infection when it was killed.

Horse No. 11.—Native pony; contracted the disease naturally. No information as to length of time the animal had been infected. Œdema of the abdomen and other symptoms of surra present. This animal was not treated and died on June 1 of surra, the animal being used for control purposes.

Horse No. 12.—Arrived in the advanced stages of surra. The animal was killed on May 12, but no trypanosomata were found in the spleen, bone marrow, lymphatic glands, or blood. Fluid from the lateral ventricle of the brain was collected and centrifugated. A few trypanosomata were found in the sediment.

Horse No 13.—Large American horse; infected artificially with 1 cubic centimeter of the blood of a monkey infected with surra. This monkey had been infected six days previously with the blood of horse No. 8. Five days later the blood of the horse showed numerous trypanosomata. This animal was untreated and died of surra on July 14, less than one month from the date of its infection.

The course of the disease in this animal proved the virulence of the strain employed in the further experiments.

Horse No. 14.—American horse (weight 431 kilos); infected artificially on July 12 by subcutaneous injection of 10 cubic centimeters of the blood of horse No. 13. July 21 numerous trypanosomata were present in the animal's blood. On July 22 an injection of 28 grams of arsenophenylglycin in 300 cubic centimeters of salt solution was given intravenously (0.065 gram per kilo). The trypanosomata disappeared from the blood and did not reappear. The animal was found dead on August 15. The cause of death in this instance is obscure, but was probably due to the toxic effect of the drug. The horse died quite suddenly, having appeared to be in good condition on the previous day, when he suddenly fell to the ground.

Horse No. 15.—American horse (weight 375 kilos); infected artificially with 2 cubic centimeters of the blood of horse No. 14, containing numerous trypanosomata. Five days later parasites were present in the blood of the animal and the following day they were numerous. On this date the animal was given 25 grams of arsenophenylglycin intravenously (0.066 gram per kilo). On July 29, two days later, this animal died of arsenic poisoning.

Horse No. 16.—American horse (weight 502 kilos); infected with 2 cubic centimeters of the blood of horse No. 14 on July 22. On July 26 the blood of this animal was positive for trypanosomata. On July 29, 11.5 grams of arsenophenylglycin were injected intravenously (0.05 gram per kilo). The parasites were absent from the blood of the animal on August 3. Although the blood was negative for parasites, the animal appeared sick and did not eat well. For this reason a second injection of 10 grams of arsenophenylglycin was given intravenously. Following this injection the horse suddenly became weak, staggered and fell to the ground. The animal died on August 9 of arsenic poisoning.

Horse No. 17.—American horse (weight 460.4 kilos); infected with 2 cubic centimeters of the blood of horse No. 16 on July 29. On August 2, blood positive for trypanosomata. On August 7, 23 grams of arsenophenylglycin were injected intravenously (0.05 gram per kilo). The trypanosomata disappeared from the blood and remained absent until August 30. On August 30 the blood was positive for trypanosomata and the animal was killed.

Horse No. 18.—American horse (weight 466.8 kilos); infected artificially with 5 cubic centimeters of the blood of horse No. 17. On August 10 trypanosomata were present in the blood of this animal and it was treated with 1,250 cubic centimeters of 0.5 per cent potassium antimonyl tartrate to which hydrogen sulphide had been added and then carbon dioxide had been passed through the solution to remove the excess of hydrogen sulphide. The trypanosomata disappeared from the blood and remained absent for fifteen days when they again reappeared in large numbers. On the same day 1,500 cubic centimeters of the same solution were injected. The animal was found dead on the following day, death being due to antimony poisoning.

Horse No. 19.—Native horse admitted on October 18, having acquired the surra infection naturally. Numerous trypanosomata were present in the blood. On October 27, when the disease was well advanced the animal was killed. On October 19, 5 cubic centimeters of this animal's blood, which contained numerous trypanosomata, were injected into horse No. 20.

Horse No. 20.—Infected with 5 cubic centimeters of the blood of horse No. 19. On October 27 trypanosomata were numerous in the blood, when 30 grams of arsenophenylglycin were injected intravenously (0.052 gram per kilo). Following the inoculation the parasites disappeared from the circulation. Although the animal's condition appeared good, after the injection diarrhœa developed and

the animal was found dead on the morning of November 4, evidently having succumbed to arsenic poisoning.

Horse No. 21.—American horse (weight 521 kilos); injected with 5 cubic centimeters of the blood of horse No. 19 on October 19. October 27 positive for trypanosomata. Thirty grams of arsenophenylglycin were given by subcutaneous and intravenous injection (0.058 gram per kilo). There was profuse perspiration following the injection and the animal became very restless. On November 6 a considerable area of induration appeared just above the front shoulders of the horse. This was incised but no pus was present. On November 12, 10 cubic centimeters of the blood of the horse were injected into a monkey. This monkey later developed surra on November 19, from which it died on November 24. On November 12, although the condition of the animal appeared good, 20 grams of arsenophenylglycin were again injected intravenously. On November 29, although the blood of the horse was negative for parasites by microscopical examination, a second monkey was inoculated with its blood. This animal developed surra infection on December 6. On November 29, the horse was given 19 grams of arsenophenylglycin intravenously. On December 7 the blood of the horse was again injected into another monkey. This animal remained negative for parasites. December 7 the horse was given another injection of 20 grams of arsenophenylglycin intravenously.

On January 3 the temperature registered 40°. An examination of the blood showed a few trypanosomata present. Twenty-five grams of the drug were then given intravenously. The parasites then disappeared from the blood and were not found present on repeated examinations up to January 20. On January 19 the condition of the horse appeared to be good. On January 20, although his temperature was normal, he refused to eat and became unable to rise and died a few hours later. The autopsy showed advanced glomerular nephritis, cloudy swelling of the liver and heart muscle. The large intestine was greatly distended with gas. The spleen was somewhat soft and friable. Microscopic examination of the blood was negative after repeated examinations. The cerebral fluid from the ventricles and the spinal fluid were centrifugated and one trypanosoma was found in each of three smears made from the sediment. A monkey was then inoculated with some of the blood and later developed surra.

The treatment of this horse demonstrates very forcibly the inefficiency of the drug in some instances. This animal had received five intravenous injections of arsenophenylglycin in amounts of 30, 20, 19, 20 and 25 grams. Notwithstanding the fact that at autopsy marked lesions due to arsenic poisoning were present, nevertheless a few trypanosomata had resisted the action of the drug and these were active and proved capable of causing infection and death in another animal. Furthermore, the treatment was begun in this horse eight days after infection.

Horse No. 25.—American horse, brought to laboratory on February 1 for treatment for surra. At the time of its arrival the disease was evidently well advanced. A blood examination showed the presence of trypanosomata and of microfilariæ. Nineteen grams of arsenophenylglycin were injected intravenously. On the following day there was considerable swelling about the point of the injection. The animal gradually grew worse, the œdema increased, and it died on February 15. Three days before its death there was considerable fever, but the blood was negative for trypanosomata.

Mule (American) No. 26.—This animal was observed in the Province of Bulacan. The blood was examined on December 19 and trypanosomata found therein.

On this date 19 grams of arsenophenylglycin were injected intravenously. The animal was brought to Manila two days later. At this time there were no evident symptoms of surra. On December 27, 20 grams of arsenophenylglycin were again injected intravenously. On January 6, 23 grams of the drug were given intravenously. The blood remained negative, although almost daily examinations of it were performed, until February 2, when a few trypanosomata were found present. On February 2, 25 grams of arsenophenylglycin were injected intravenously. A monkey was inoculated on this date with the blood of this mule and it later developed surra. The mule died on February 9, the treatment having evidently proved unsuccessful.

Surra in Bulacan Province.—A small outbreak of surra recently occurred in Bulacan Province, near Manila, and a few of the infected animals were treated there. The treated animals were all kept and worked together. Consequently, when a relapse of the infection occurred in one of them the others were exposed. It was practicable to visit the animals for examination and treatment only once a week. Owing to the favorable results that had been reported by Holmes in India from the treatment of trypanosomiasis in horses by a combination of atoxyl and orpiment, we determined to give this method, also, a trial in these animals. Seven mules and 3 horses were treated in a somewhat similar manner, as may be seen from the notes of the experiments which follow. In some instances arsacetin or arsenophenylglycin was substituted for the atoxyl.

Five mules were given alternate doses of atoxyl and orpiment, the atoxyl being given subcutaneously and the orpiment (arsenic trisulphide) by mouth. In all but one of these, relapses occurred shortly after the treatment was stopped, and we deemed it advisable to substitute arsenophenylglycin for this method of treatment. The details of these experiments are as follows:

Horse No. 22.—Native horse; contracted surra naturally. Length of time infected unknown. Numerous trypanosomata in the blood at the time of entrance. On December 4, 5 grams of arsacetin were given subcutaneously. On December 5 no trypanosomata were found. On December 10, 10 grams of arsenic sulphide were administered by mouth. Following this, the horse became unable to rise and died of arsenic poisoning on December 8. The arsenic sulphide used in this case was precipitated and was not thoroughly washed with alcohol or ether and probably contained a percentage of white arsenic.

Horse No. 23.—American horse infected with 5 cubic centimeters of the blood of horse No. 22 on December 3. On December 7 its blood was positive for trypanosomata. The parasites became numerous on December 9, on which date 15 grams of arsenic sulphide were administered in capsule by mouth. On December 10 the parasites were still numerous; December 11, only one trypanosoma was found in a drop of fresh blood. On the same date 5 grams of arsacetin were injected subcutaneously. On December 13 another dose of 10 grams of arsenic sulphide was given. December 16, 5 grams of atoxyl; December 23, 10 grams of arsenic sulphide; December 27, 5 grams of atoxyl; January 2, 20 grams of arsenic sulphide; January 5, 5 grams of atoxyl; January 9, 20 grams of arsenic sulphide; January 13, 5 grams of atoxyl; January 17, 25 grams of arsenic sulphide.

On this date 10 grams of arsenophenylglycin were injected intravenously. The animal was brought to Manila two days later. At this time there were no evident symptoms of surra. On December 27, 20 grams of arsenophenylglycin were again injected intravenously. On January 6, 23 grams of the drug were given intravenously. The blood remained negative, although almost daily examinations of it were performed, until February 2, when a few trypanosomata were found present. On February 2, 25 grams of arsenophenylglycin were injected intravenously. A monkey was inoculated on this date with the blood of this mule and it later developed surra. The mule died on February 9, the treatment having evidently proved unsuccessful.

Surra in Bulacan Province.—A small outbreak of surra recently occurred in Bulacan Province, near Manila, and a few of the infected animals were treated there. The treated animals were all kept and worked together. Consequently, when a relapse of the infection occurred in one of them the others were exposed. It was practicable to visit the animals for examination and treatment only once a week. Owing to the favorable results that had been reported by Holmes in India from the treatment of trypanosomiasis in horses by a combination of atoxyl and orpiment, we determined to give this method, also, a trial in these animals. Seven mules and 3 horses were treated in a somewhat similar manner, as may be seen from the notes of the experiments which follow. In some instances arsacetin or arsenophenylglycin was substituted for the atoxyl.

Five mules were given alternate doses of atoxyl and orpiment, the atoxyl being given subcutaneously and the orpiment (arsenic trisulphide) by mouth. In all but one of these, relapses occurred shortly after the treatment was stopped, and we deemed it advisable to substitute arsenophenylglycin for this method of treatment. The details of these experiments are as follows:

Horse No. 22.—Native horse; contracted surra naturally. Length of time infected unknown. Numerous trypanosomata in the blood at the time of entrance. On December 4, 5 grams of arsacetin were given subcutaneously. On December 5 no trypanosomata were found. On December 10, 10 grams of arsenic sulphide were administered by mouth. Following this, the horse became unable to rise and died of arsenic poisoning on December 8. The arsenic sulphide used in this case was precipitated and was not thoroughly washed with alcohol or ether and probably contained a percentage of white arsenic.

Horse No. 23.—American horse infected with 5 cubic centimeters of the blood of horse No. 22 on December 3. On December 7 its blood was positive for trypanosomata. The parasites became numerous on December 9, on which date 15 grams of arsenic sulphide were administered in capsule by mouth. On December 10 the parasites were still numerous; December 11, only one trypanosoma was found in a drop of fresh blood. On the same date 5 grams of arsacetin were injected subcutaneously. On December 13 another dose of 10 grams of arsenic sulphide was given. December 16, 5 grams of atoxyl; December 23, 10 grams of arsenic sulphide; December 27, 5 grams of atoxyl; January 2, 20 grams of arsenic sulphide; January 5, 5 grams of atoxyl; January 9, 20 grams of arsenic sulphide; January 13, 5 grams of atoxyl; January 17, 25 grams of arsenic sulphide.

This animal is apparently well. Repeated examinations of its blood and inoculations of it into 3 monkeys have been made. None of the animals have developed trypanosomiasis.

Horse No. 24.—American colt infected artificially with 5 cubic centimeters of the blood of horse No. 22 on December 4. December 7 blood examination showed a few trypanosomata. December 9, trypanosomata numerous, when 10 grams of arsenic sulphide were administered by the mouth in capsules. December 10, trypanosomata numerous; December 11, 5 grams of arsacetin injected subcutaneously. From December 11 to December 17 the horse was given alternate doses of arsenic sulphide and arsacetin and died on December 18 of arsenic poisoning.

Mule (American) No. 27.—This animal was also examined at Bulacan on December 17 and trypanosomata found in its blood. On the same date 15 grams of arsenic sulphide were given by mouth. December 19 the blood was still positive for parasites and 15 grams of arsenophenylglycin were injected intravenously. The animal was brought to the laboratory on December 21. Its temperature on examination was 40° and there was considerable œdema between the front shoulders. The temperature dropped to normal on December 24, although the marked œdema increased along the whole of the abdomen. December 27, 20 grams of arsenophenylglycin were given intravenously, although the blood remained negative for parasites. Twenty-five grams of the drug were again given on January 4 and 25 grams on January 25. A monkey was inoculated on January 25 just before the injection of the drug and did not develop surra infection. On February 14, 10 grams of arsenic sulphide were given the animal by the mouth, through a mistake on the part of the attendant. On March 23 trypanosomata reappeared in his blood and he was given 30 grams of arsenophenylglycin.

This animal is at present alive and its blood contains no trypanosomata. It seems not unlikely that this animal was reinfected with surra from another mule which was suffering with the disease and which was kept with it.

Mule (American) No. 28.—This mule was found positive for trypanosomata in Bulacan on December 17. On December 19, 15 grams of arsenophenylglycin were injected intravenously. Shortly after this injection the temperature which had been elevated became normal. On December 27, 15 grams of arsenophenylglycin were given intravenously. The blood remained negative until January 8, when trypanosomata reappeared. The animal was then inoculated with 5 grams of atoxyl, but did not bear the inoculation well and three days later was in a very weak condition. On January 14 the blood examination showed no parasites. On January 22, 24, and 26, 10 grams of arsenic sulphide were given by mouth and on January 29, 5 grams of atoxyl subcutaneously. On January 31, 20 grams of arsenic sulphide; February 2, 30 grams of arsenic sulphide; February 5, 5 grams of atoxyl; February 7, 20 grams of arsenic sulphide; February 9, 20 grams of arsenic sulphide. February 14 the animal became still weaker. He developed pressure sores, and on March 19 trypanosomata reappeared in his blood and he was destroyed.

Mule (American) No. 29.—This animal was also observed at Bulacan. It appeared in excellent condition at the time the trypanosomata were found on December 17, on which date 15 grams of arsenic sulphide were given by mouth. Decembr 19 the blood was still positive for parasites and 5 grams of atoxyl were injected subcutaneously. December 21, 20 grams of arsenic sulphide were given.

December 23 the blood was negative for parasites and 5 grams of atoxyl were injected; December 25, 30 grams of arsenic sulphide; December 28, 5 grams of atoxyl; January 1, 15 grams of arsenic sulphide. January 4 trypanosomata reappeared in the blood and 5 grams of atoxyl were given; January 6, 20 grams of arsenic sulphide; January 8, 5 grams of atoxyl; January 10, 20 grams of arsenic sulphide; January 14, 5 grams of atoxyl; January 18, 20 grams of arsenic sulphide; January 22, 5 grams of atoxyl; January 24, 20 grams of arsenic sulphide; January 26, 20 grams of arsenic sulphide; January 29, 5 grams of atoxyl; January 31, 20 grams of arsenic sulphide; February 3, 20 grams arsenic sulphide; February 5, 5 grams of atoxyl; February 7, 20 grams of arsenic sulphide; February 9, 20 grams arsenic sulphide. February 14, although the condition of the animal appeared good, trypanosomata reappeared in the blood. February 14, 5 grams of atoxyl; February 17, 25 grams arsenic sulphide; February 19, 25 grams of arsenophenylglycin. Since this date the animal has been negative for trypanosomata and is in good condition.

Mule (American) No. 30.—Observed at Bulacan December 17. Examination of the blood positive for trypanosomata. Fifteen grams of arsenic sulphide given by mouth; December 19, 5 grams of atoxyl; December 21, 20 grams arsenic sulphide; December 23, 5 grams atoxyl; December 25, 20 grams arsenic sulphide; December 28, 5 grams of atoxyl. December 30 the animal suffered from diarrhœa and no treatment was given; January 4, 15 grams of arsenic sulphide. January 8 trypanosomata reappeared in the blood. The animal was given 5 grams of atoxyl; January 12, 20 grams arsenic sulphide. Diarrhœa and colic developed. January 14, 5 grams of atoxyl; January 18, 20 grams arsenic sulphide; January 22, 5 grams atoxyl; January 24, 20 grams arsenic sulphide; January 26, 20 grams arsenic sulphide; January 29, 5 grams atoxyl; January 31, 20 grams arsenic sulphide; February 3, 20 grams arsenic sulphide; February 5, 5 grams of atoxyl; February 7, 20 grams arsenic sulphide; February 9, 20 grams arsenic sulphide; February 14, 5 grams atoxyl; February 17, 20 grams arsenic sulphide; February 19, 20 grams arsenophenylglycin. March 23 trypanosomata reappeared in the blood, when 25 grams of arsenophenylglycin were injected intravenously. The animal is still alive and free from parasites.

Mule (American) No. 31.—Found positive for surra on microscopic examination on December 23 at Bulacan. December 23, 5 grams of atoxyl; December 25, 20 grams arsenic sulphide; December 28, 5 grams atoxyl; December 30, 25 grams arsenic sulphide; January 1, 5 grams of atoxyl; January 4, 20 grams of arsenic sulphide; January 6, 20 grams of arsenic sulphide; January 8, 5 grams atoxyl; January 10, 20 grams arsenic sulphide; January 12, 20 grams arsenic sulphide; January 14, 5 grams atoxyl; January 17, 20 grams arsenic sulphide; January 22, 5 grams atoxyl. The animal having been treated about one month, treatment was discontinued until February 14. During this time the blood was negative. February 14, the blood was found positive for trypanosomata, when 5 grams of atoxyl were given; February 17, 20 grams arsenic sulphide; February 19, 20 grams arsenophenylglycin. The blood has since been negative.

Mule No. 32.—American mule found positive for surra by microscopic examination on December 19 at Bulacan, when 5 grams of atoxyl were given; December 21, 20 grams of arsenic sulphide; December 23, blood positive, 5 grams of atoxyl; December 25, 20 grams arsenic sulphide; December 28, 5 grams of atoxyl; December 30, 5 grams atoxyl; January 1, 20 grams arsenic sulphide; January 4, 5 grams atoxyl; January 6, 20 grams arsenic sulphide; January 8, 5 grams atoxyl; January 10, 20 grams arsenic sulphide; January 12, 20 grams arsenic sulphide; January 14, 5 grams atoxyl; January 17, 20 grams arsenic sulphide; January 22, 5 grams atoxyl. Treatment was then stopped, the animal

December 23 the blood was negative for parasites and 5 grams of atoxyl were injected; December 25, 30 grams of arsenic sulphide; December 28, 5 grams of atoxyl; January 1, 15 grams of arsenic sulphide. January 4 trypanosomata reappeared in the blood and 5 grams of atoxyl were given; January 6, 20 grams of arsenic sulphide; January 8, 5 grams of atoxyl; January 10, 20 grams of arsenic sulphide; January 14, 5 grams of atoxyl; January 18, 20 grams of arsenic sulphide; January 22, 5 grams of atoxyl; January 24, 20 grams of arsenic sulphide; January 26, 20 grams of arsenic sulphide; January 29, 5 grams of atoxyl; January 31, 20 grams of arsenic sulphide; February 3, 20 grams arsenic sulphide; February 5, 5 grams of atoxyl; February 7, 20 grams of arsenic sulphide; February 9, 20 grams arsenic sulphide. February 14, although the condition of the animal appeared good, trypanosomata reappeared in the blood. February 14, 5 grams of atoxyl; February 17, 25 grams arsenic sulphide; February 19, 25 grams of arsenophenylglycin. Since this date the animal has been negative for trypanosomata and is in good condition.

Mule (American) No. 30.—Observed at Bulacan December 17. Examination of the blood positive for trypanosomata. Fifteen grams of arsenic sulphide given by mouth; December 19, 5 grams of atoxyl; December 21, 20 grams arsenic sulphide; December 23, 5 grams atoxyl; December 25, 20 grams arsenic sulphide; December 28, 5 grams of atoxyl. December 30 the animal suffered from diarrhœa and no treatment was given; January 4, 15 grams of arsenic sulphide. January 8 trypanosomata reappeared in the blood. The animal was given 5 grams of atoxyl; January 12, 20 grams arsenic sulphide. Diarrhœa and colic developed. January 14, 5 grams of atoxyl; January 18, 20 grams arsenic sulphide; January 22, 5 grams atoxyl; January 24, 20 grams arsenic sulphide; January 26, 20 grams arsenic sulphide; January 29, 5 grams atoxyl; January 31, 20 grams arsenic sulphide; February 3, 20 grams arsenic sulphide; February 5, 5 grams of atoxyl; February 7, 20 grams arsenic sulphide; February 9, 20 grams arsenic sulphide; February 14, 5 grams atoxyl; February 17, 20 grams arsenic sulphide; February 19, 20 grams arsenophenylglycin. March 23 trypanosomata reappeared in the blood, when 25 grams of arsenophenylglycin were injected intravenously. The animal is still alive and free from parasites.

Mule (American) No. 31.—Found positive for surra on microscopic examination on December 23 at Bulacan. December 23, 5 grams of atoxyl; December 25, 20 grams arsenic sulphide; December 28, 5 grams atoxyl; December 30, 25 grams arsenic sulphide; January 1, 5 grams of atoxyl; January 4, 20 grams of arsenic sulphide; January 6, 20 grams of arsenic sulphide; January 8, 5 grams atoxyl; January 10, 20 grams arsenic sulphide; January 12, 20 grams arsenic sulphide; January 14, 5 grams atoxyl; January 17, 20 grams arsenic sulphide; January 22, 5 grams atoxyl. The animal having been treated about one month, treatment was discontinued until February 14. During this time the blood was negative. February 14, the blood was found positive for trypanosomata, when 5 grams of atoxyl were given; February 17, 20 grams arsenic sulphide; February 19, 20 grams arsenophenylglycin. The blood has since been negative.

Mule No. 32.—American mule found positive for surra by microscopic examination on December 19 at Bulacan, when 5 grams of atoxyl were given; December 21, 20 grams of arsenic sulphide; December 23, blood positive, 5 grams of atoxyl; December 25, 20 grams arsenic sulphide; December 28, 5 grams of atoxyl; December 30, 5 grams atoxyl; January 1, 20 grams arsenic sulphide; January 4, 5 grams atoxyl; January 6, 20 grams arsenic sulphide; January 8, 5 grams atoxyl; January 10, 20 grams arsenic sulphide; January 12, 20 grams arsenic sulphide; January 14, 5 grams atoxyl; January 17, 20 grams arsenic sulphide; January 22, 5 grams atoxyl. Treatment was then stopped, the animal

having been treated for one month. On March 5, trypanosomata reappeared in the blood. March 9, 24 grams of arsenophenylglycin were given intravenously. The animal died four days later.

Mule (American) No. 33.—Found positive for surra on December 28 at Bulacan. Arsenic sulphide 20 grams by mouth. December 30, blood negative, arsenic sulphide 20 grams. Animal found dead on morning of January 1.

Epidemic at Alabang.—During the past year an epidemic of surra was also observed among the horses of the Government stock farm at Alabang. On September 3 we visited Alabang and found 17 horses infected with surra. The blood in each instance was examined and the trypanosomata found therein. In many of the animals the parasites were very numerous and the horses showed well-marked symptoms of surra. In the following few weeks 3 other horses were found infected with surra at Alabang and these were treated with the others. The horses, during the course of their treatment, were kept in an open field at one corner of the farm and were fed under a *nipa* roof, which afforded some protection from the sun. Flies were abundant, to the bites of which the horses were naturally exposed. The conditions, therefore, were such as might be met with in combating the disease in provincial districts with no protection against reinfection, either from biting flies or from contact with other infected animals.

Therefore, the conditions surrounding the experiment seemed most unfavorable for obtaining good results from treatment.

The injections of arsenophenylglycin were given intravenously in approximately 10 per cent solution. Since no facilities were at hand for holding the horses while they were being treated, it frequently happened that during the inoculation the needle escaped from the vein so that some of the solution was given subcutaneously. When this occurred, severe local reaction sometimes followed which resulted in abscess formation. However, the abcesses healed later without trouble. No animals apparently died from the local effect of the drug. Owing to the conditions just described, it was impossible in the course of the experiments to determine whether a reappearance of the trypanosomata in the blood of individual horses indicated a relapse, or a reinfection. The experiments in monkeys indicated that a single large dose of the drug gave the best results. However, owing to the fact that some of our horses would almost certainly have been reinfected with surra during the period following the first treatment, we felt compelled to repeat the dose at intervals, even though the blood remained negative in most of the cases. Our experiments with horses have shown us that as few injections as possible should be made, owing to the danger of the production by the drug of grave lesions in the internal organs. Table III gives the results of the treatment of these horses.

Discussion of Table III.—During the few weeks following the first treatment of the horses, trypanosomata reappeared in the blood of a number of them, and since we did not have a sufficient quantity of arseno-

phenylglycin on hand to continue the treatment of all of the animals, some were destroyed. A few other horses died from the toxic effects of the drug. There remained, then, on November 19 only 8 horses alive. Four of these were brought to Manila and placed in screened stalls. These latter have all remained entirely free from parasites for six months [2] and are in excellent condition at the present time. The blood of these horses has been subjected to repeated examination and as large an amount as 20 cubic centimeters has been repeatedly inoculated at intervals into monkeys with negative results.

One of the horses, No. 73, had a slight rise in temperature on December 13; fearing a relapse, this horse's blood was at once injected into a monkey and the horse was then given a full dose of arsenophenylglycin. The monkey, however, remained free from trypanosomata, and we feel justified in concluding that the rise in temperature was due to some other cause than trypanosomiasis. These horses, we believe, have been cured of surra. One of them has been kept at work for over a month at Manila. Of the other horses, 2 have shown a reappearance of trypanosomata in the peripheral blood after having been free from parasites for three months and five months, respectively. In view of the fact that the horses which were brought to Manila and kept in screened stalls have not shown any relapses, and, also, because the 4 horses left at Alabang remained in the same corner of the farm where the horses infected with surra were originally segregated, it seems not unlikely that the reappearance of the trypanosomata in the two instances just cited resulted from a reinfection from biting insects and was not due to a relapse of the original infection. However, it is possible that a relapse may have occurred. Both of these horses were subjected to further treatment; one succumbed a day or so after the administration of a large dose of arsenophenylglycin, the other is still alive and in good condition three months after the last treatment.

The other horses at Alabang, although negative for trypanosomata, were treated on November 19 as a prophylactic measure because of their intimate contact with the horse showing trypanosomata in its blood. They have remained negative for trypanosomata and are in excellent condition at the present time. They have not been under daily observation, as have the horses that were brought to Manila, but we are inclined to regard them as free from the disease.

We believe, then, that we have on hand 7 horses, all in good condition, which have been definitely and permanently cured of surra by the intravenous administration of arsenophenylglycin.

Surra runs a very chronic course in cattle in the Philippine Islands, the animals harboring the parasites over long periods of time, during which their general health may remain unimpaired. This renders it

[2] At the time of the reading of this proof, now 9 months.

extremely difficult to determine whether or not a drug used in the treatment of these animals is efficacious. Therefore, although we have treated a number of bullocks that were infected with the disease, we prefer to express no opinion at this time with regard to the action of the drug on these animals.

SUMMARY.

In the use of arsenophenylglycin two methods of treatment may be considered, as already outlined by Ehrlich: First, that by stages in which relatively small doses are given and at repeated intervals; second, the treatment by one or several large doses. In monkeys and horses there is no question but that the second method of treatment is the more favorable one. In horses, the most satisfactory results have been obtained where we have given an amount probably very close to the fatal dose to the animal infected. Unfortunately, in horses the margin between the dose necessary to effect a complete cure and the fatal one seems very small (see horse No. 21), too small for us to be able to determine the amount most favorable for any given case. Moreover, the susceptibility of horses to the effect of the drug naturally varies to some extent. The condition under which the animal is kept after treatment, whether favorable or otherwise, may possibly be the deciding point as to whether the animal recovers or dies. Each repetition of a large dose obviously exposes the animal to increased danger of poisoning, while the opportunity of producing a cure is diminished. These facts are evident upon consideration of the action of atoxyl in the human body and from the results of our own experiments.

In conclusion, we do not hesitate to say that arsenophenylglycin has proved to be by far the most satisfactory means of treatment of trypanosomiasis yet discovered. The drug has shown itself to be very efficacious for the treatment and cure of surra infection in monkeys. However, while the results in horses, are the best that have as yet been obtained, they are not nearly as encouraging as in monkeys. We have, however, for the first time been able to cure horses afflicted with surra, and this we never could accomplish before by any other means. While it appears that we can save a certain percentage of the horses infected with surra during an epidemic, we can never predict with certainty whether in a given instance we will be able to cure the horse, or whether he will succumb first to the action of the drug. However, since, the disease is invariably fatal in these animals without treatment, a trial should be made in the case of every horse of any great value. In the epidemic at Alabang, we were able to save a sufficient percentage of the horses to demonstrate that the means is of some practical value in the treatment of surra during an epidemic.

It is not our intention in the present paper to report upon the results we have obtained in the treatment of trypanosomiasis with a large number of other compounds prepared by Doctor Oechslin, of the chemical

laboratory of this Bureau. This work is still being continued at the present time, but so far, no results as satisfactory as those with arsenophenylglycin have yet been obtained.

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TABLE I.—*Dosage of arsenophenylglycin in normal monkeys.*

Weight of monkey in grams.	Dose of arsenophenylglycin in grams per kilo.	Death after—	Weight of monkey in grams.	Dose of arsenophenylglycin in grams per kilo.	Death after—	Weight of monkey in grams.	Dose of arsenophenylglycin in grams per kilo.	Death after—
2,250	0.10		2,100	0.18		1,520	0.26	3 days.
1,800	0.10		2,350	0.20		1,440	0.28	
2,350	0.12		2,170	0.20		2,170	0.30	3 days.
2,300	0.12		2,820	0.22		2,040	0.30	8 days.
2,300	0.14		2,700	0.22	6 days.	2,370	0.32	3 days.
2,320	0.14		2,620	0.24		1,970	0.32	2 days.
1,900	0.16		2,350	0.24		1,730	0.34	10 days.
2,440	0.16		2,300	0.26		1,750	0.36	2 days.
2,300	0.18		2,400	0.26	3 days.			

TABLE II, SERIES I (Monkeys).

No. of monkey.	Weight in grams.	First treatment in grams per kilo.	Interval between doses.		Second treatment in grams per kilo.	Result.	Period free from trypanosomata.
			Days.	Blood examination.			
4477	2,500	0.04	18	Pos.	0.08	Alive	7 months.
4432	850	0.04	28	Pos.	0.08		2½ months.[a]
4332	1,620	0.04				Died in 8 days (negative)	
4473	3,200	0.05	20	Pos.	0.08	Died in 4 days	
4479	1,100	0.05	34	Pos.	0.08	Died after 3 months	
4341	1,520	0.05				Died 5 days	
4472	5,250	0.06	27	Pos.	0.08	Died in 6½ months	
4476	1,950	0.06	19	Pos.	0.08	Alive	7 months.
4421	1,220	0.06				Died in 55 days (positive)	
4293	Died 3 days after inoculation with surra—not treated						
4474	5,900	0.07				Alive	8½ months.
4480	1,550	0.07				do	Do.
4337	1,770	0.07	34	Pos.	0.09	Died in 19 days (negative)	
4475	3,950	0.08				Alive	Do.
4481	1,600	0.08				do	Do.
4419	1,200	0.08	23	Pos.	0.10	do	6 months.
4420	750	0.09	51	Pos.	0.10	Died in 3 hours	
4350	1,900	0.09				Died in 17 days (negative)	
4478	2,500	Control—not treated				Died 17 days after inoculation	
4422	850	Control—not treated				Died 14 days after inoculation	

[a] This monkey died 10 days after a third treatment with a dose of 0.15 gram per kilo.

TABLE II, SERIES II (Monkeys).

No. of monkey.	Weight at first treatment.	First dose of arsenophenylglycin in grams per kilo.	Interval between doses.		Second dose of arsenophenylglycin in grams per kilo.	Result.	Period free from trypanosomata.
			Days.	Blood examination.			
4559	1,250	0.08	31	Pos.	0.1	Died in 1 day	
4560		0.08	31	Neg.	0.1	Died in 17 days	
4561	2,700	0.08				Alive	7 months.
4562	1,400	0.02	[a]4	Neg.	0.1	Died in 4 days	
4563	Control—not treated.					Died in 26 days	
4564	2,700	0.08	[a]2	Neg.	0.15	Alive	Do.
4565		0.08				Lost	3½ months.
4566		0.80	[a]4	Neg.	0.15	Died in 2 days	

[a] Months.

TABLE II, SERIES III (Monkeys).

No. of monkey.	Weight at first treatment.	First dose of arsenophenylglycin in grams per kilo.	Result.	Period free from trypanosomata.
4612	2,250	0.1	Alive	5⅔ months.
4613	1,850	0.1	Died in 16 days	
4614	1,700	0.12	Alive	Do.
4615	2,350	0.14	do	Do.
4616	2,200	0.14	Lost	4½ months.
4617	2,320	0.14	Died in 22 days (negative)	
4618	1,500	0.16	Died in 5 days	
4619	2,450	0.16	Alive	5⅔ months.
4620	2,250	0.18	do	Do.
4621	2,050	0.18	do	Do.
4622	2,300	0.20	do	Do.
4623	2,150	0.20	do	Do.
4624	2,950	0.22	do	Do.
4625	2,000	0.22	Died in 3 months (negative)	
4626	2,170	0.24	Alive	Do.
4627	2,250	0.24	Died in 13 days	
4628	2,000	0.26	Died in 10 days	
4629	2,010	0.26	Alive	Do.
4630	Control—not treated		Died 5 days after inoculation	
4631	Control—not treated		Died 33 days after inoculation	
4632	Control—not treated		Died 22 days after inoculation	

TABLE II, SERIES IV (Monkeys).

No. of monkey.	Weight at first treatment.	First dose of arsenophenylglycin in grams per kilo.	Result.	Period free from trypanosomata.
4655	1,570	0.26	Died in 12 days	
4654	1,450	0.28	Alive	5 months.
4656	2,250	0.30	Died in 1 day	
4657	1,950	0.30	do	
4658	2,150	0.32	Alive	Do.
4659	1,250	0.32	do	Do.
4660	1,700	0.34	Died in 8 days	
4661	1,350	0.36	Died in 1 day	
4658	Control—not treated.		Died 17 days after inoculation.	

TABLE III (Horses).

No. of horse.	Age.	First treatment.		Interval.		Second treatment.		Interval.		Third treatment.		Interval.		Fourth treatment.		Interval.		Fifth treatment.		Interval.		Sixth treatment.		Entire period free from trypanosomata.	March 5 (period since last treatment).
		Date.	Dose.	Days.	Blood examination.[a]	Date.	Dose.	Days.	Blood examination.[a]	Date.	Dose.	Days.	Blood examination.[a]	Date.	Dose.	Days.	Blood examination.[a]	Date.	Dose.	Days.	Blood examination.[a]	Date.	Dose.		
	Y. m.		*G.*				*G.*				*G.*				*G.*				*G.*				*G.*		
2	1 2	Sept. 3	5	13	N	Sept. 16	8	8	N	Sept. 21	10	18	P												
29	2 6	Sept. 3	13	17	P	Sept. 20	[b]																		
37	10	Sept. 3	20	13	N	Sept. 16	25	5	N	Sept. 21	[c]10	25	N	Oct. 16	[c]10	34	N	Nov. 19	20	82	P	Feb. 9	25	15 months[d]	
37A	10	Sept. 3	5	13	P	Sept. 16	10	5	N	Sept. 21	[c]10	25	N	Oct. 16	[c]8	34	N	Nov. 19	[c]10					5½ months.	3½ months.
38	7	Sept. 3	30	18	N	Sept. 21	30																	6 months.	5½ months.
43	8	Sept. 3	20	18	N	Sept. 21	30	20	P	[e]															
58	2 5	Sept. 3	15	17	P	Sept. 20	[b]																		
61	2 5	Sept. 3	18	18	N	Sept. 21	[c]10																	6 months.	6 months.
65	2 5	Sept. 3	15	18	N	Sept. 21	[c]10	59	N	Nov. 19	15	45	P	Dec. 3	20	68	N	Feb. 9	15					3 months.	24 days.
67	2 9	Sept. 3	24	13	N	Sept. 16	25	5	N	Sept. 21	19	25	N	Oct. 16	15									6 months.	5 months.
73	1 4	Sept. 3	8	13	P	Sept. 16	10	5	N	Sept. 21	[c]8	73	N	Dec. 13	15									5½ months.	82 days.
75	12	Sept. 3	25	18	N	Sept. 21	35	(c)																	
76	12	Sept. 3	24	(f)																					
78	9	Sept. 3	30	18	N	Sept. 21	[c]10	25	N	Oct. 16	[c]15	34	N	Nov. 19	20									6 months.	3½ months.
88	1 7	Sept. 3	13	13	N	Sept. 16	15	5	N	Sept. 21	15	25	N	Oct. 16	15	(g)									
96	1 6	Sept. 3	10	18	N	Sept. 21	12	24	P	Oct. 15	[b]														
49	10	Sept. 16	25	5	N	Sept. 21	[c]10	[b]																	
5	3 6	Sept. 16	10	5	N	Sept. 21	[c]8	[h]																	
74	1 9	Sept. 24	[c]10	[i]																					
69	1 9	Sept. 3	8	18	P	Sept. 21																			

[a] N=negative; P=positive.
[b] Destroyed.
[c] Arsacetin was used instead of arsenophenylglycin.
[d] Died February 11.
[e] Died November 10.
[f] Died September 11.
[g] Died October 22.
[h] Died October 1.
[i] Died September 28.
[j] Died October 3.

THE ETIOLOGY OF BERIBERI.[1]

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The suggestion of a causal relationship between the consumption of white rice and the disease beriberi was first formally made in this country by Braddon (1). The observer also drew attention to the important fact that those who consumed rice which had been parboiled before husking remained free from the disease, as did also the native Malays who consumed rice prepared by primitive methods of pounding and winnowing.

A series of observations made by the writers (2) in 1907 on two parties of laborers, under conditions which excluded or adequately controlled the operation of factors other than diet, confirmed the correctness of this view of the causation of the disease. The prior observations of Fletcher (3) and Lucy (4) in this country and of Dubruel (5) in Indo-China and the recently published observations of Ellis (6) furnish further testimony, and it may now be claimed that the theory rests on a solid basis of evidence.

The mechanism by which white rice was able to produce this result has remained obscure.

Braddon suggested that "the cause of the disorder is not indeed rice, qua rice, or as an article of diet, but diseased rice; rice with which poison derived from decay, due perhaps to some fungus, or mold, or germ, or spore, originally perhaps growing upon the husk, has become mixed during the process of milling; or upon which such fungus may have grown and such poison have been produced after decortication." Eykman (7) from experiments on fowls concluded that a definite poison exists commonly in rice and that for this poison or its effects something in the pericarp is an antidote. Dubruel believed in the ingestion of an organism associated with white rice, which organism multiplying in the body produced the disease.

Following the line of thought suggested by the poison hypothesis, researches were undertaken to determine whether, from white rices actually

[1] Read at the first biennial meeting of the Far Eastern Association of Tropical Medicine held at Manila, March 10, 1910.

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associated with outbreaks of beriberi, there could be extracted by means of various solvents any substance or substances recognizable by chemical methods as poisonous in character. These researches failed of their object, though it is admitted that the accuracy of the poison hypothesis was not thereby disproved.

Certain results which emerged from chemical analysis and histological examination of the rices turned attention to the possibility of an explanation of the course of events on an hypothesis of a defect of nutrition. That this explanation was inadequate, if dietary constituents as estimated by the ordinary analytical methods were alone considered, had been shown in the preliminary investigation.

By a series of experiments on domestic fowls, the details of which will be supplied in a later publication, it was shown that these animals when fed on various kinds of rice were sensitive to differences between them. The fowls were confined in separate compartments and were in all respects under identical conditions. The manner of arrangement of the cages is shown in Plate I, fig. 1.

By further and repeated experiments with rices known to have been associated with outbreaks of beriberi, and with controls under identical conditions fed on parboiled rice, it was established that a certain reaction in fowls might be taken as an indicator of the beriberi-producing power of a given rice when forming the staple of the diet in man. Whether the disease produced in fowls be accepted or not as analogous to beriberi in man, the validity of the arguments here advanced remains unimpaired.

Rices were available that were known to have been associated with outbreaks of beriberi, samples having been taken daily during the continuance of the preliminary inquiry in 1907; also through the courtesy of Dr. J. D. Gimblette and Dr. G. D. Freer we were enabled to procure white rice which was being consumed prior to an outbreak of beriberi among Malays at the Kuala Lumpur police depot, which outbreak ceased on changing the rice supplied to the parboiled variety. It was shown that these rices when fed to fowls constantly produced a certain disease in a large proportion of them, while parboiled rice as constantly failed to produce this result in groups under identical conditions. This disease is characterized by paralysis of the legs (Plate I, fig. 2, and Plate II, fig. 3), followed by paralysis of the wings (Plate II, fig. 4) in the more severe cases. In cases showing a moderate degree of paralysis the gait resembles very closely that seen in beriberi. The nerves of fowls suffering from this disease show typical Wallerian degeneration (Plate III, fig. 5).

It is our belief that this disease, polyneuritis gallinarum, is truly analogous to beriberi in man, similar in its etiology, in its clinical manifestations, and we have shown them to be identical in their pathologic

effects, and that its occurrence should be held as important confirmatory testimony of the connection between white rice and beriberi. It is desirable, however, to emphasize the point that the acceptance or nonacceptance of this opinion is immaterial to the argument; for this purpose the occurrence of the disease is employed only as a reaction. The fact that certain white rices when forming the staple of a diet in man produce beriberi rests on quite other testimony than that supplied by experiments on domestic fowls.

The commercial varieties of white rice are numerous, but in this country, apart from the grading as to quality, two are in common use and are known, respectively, as Siam and Rangoon.

From epidemiological considerations and from experimental evidence it appears that Siam rice is considerably more potent in its beriberi-producing powers than Rangoon rice.

The proteins, fats, carbohydrates, and ash were determined for the different varieties of rice which had been employed in the experiments, with the following percentage results calculated on dried material.

	Proteins.	Fats.	Carbohydrates.	Ash.
White rice (Siam)	9.07	0.17	90.11	0.65
White rice (Rangoon)	8.44	0.81	89.90	0.85
Parboiled rice	9.48	0.51	89.12	0.89

A comparison of these results shows that the only marked difference among the rices was in respect to fat, which was most abundant in the variety known as Rangoon, less abundant in parboiled rice, and still less abundant in Siam rice. These observations, taken in conjunction with the experimental results in fowls, excluded the possibility of an explanation of the origin of beriberi on the ground of a deficiency in fat. It will be noted that these analyses did not include an estimation of the relative proportions of the inorganic salts composing the ash, nor did they take account of the manner of combination, organic or inorganic, in which these substances originally existed in the rice grain.

By a method devised in this laboratory, sections of the various rice grains were obtained of sufficient thinness to permit the examination in detail of their histologic characters. By suitable staining methods it was shown that in parboiled rice (Plate III, fig. 7) remnants of the pericarp remained attached to the rice grain, whereas in Siam rice (Plate III, fig. 8) the pericarp and the layers subjacent to it (subpericarpal layers) had been polished away. It would appear that parboiling renders the grain tough and nonfriable, in consequence the subpericarpal layers can not be removed so readily as in the untreated grain. It was further demonstrated that the layers so retained in parboiled rice contained the

most of the aleurone and oily material present in rice grains. Rice as prepared by primitive methods (Malay rice) was similarly examined, and, as might have been expected from the pounding to which this rice had been subjected, parts of the subpericarpal layers were chipped off to a varying extent, but on the whole these layers were retained to a greater extent than is the case with white rice.

Early in the course of the experiments the observation was made that parboiled rice subjected to exhaustion with hot alcohol and thereafter carefully dried in the sun to free it from alcohol, produced when fed to fowls a disease indistinguishable from that observed in birds fed on white rice, although such parboiled rice in its original state was incapable of producing this result, however long continued.

The association of the observations referred to in the two preceding paragraphs seemed to point a way to a solution of the problem. It had been shown that white rice as prepared in the mills of this country produced the same results in fowls as white rice known to have been associated with beriberi. If, now, a substance or substances residing in the outer layers which are polished away in white rice and are retained in parboiled rice could be added to white rice and so prevent its harmful effects it was conceived that the nutritive hypothesis would thereby be supported.

In accordance with this idea the following experiments were initiated:

A rice mill in Singapore was visited and there was obtained (A) a quantity of the grain deprived of the husk; (B) a quantity of the polished rice from the same lot of grain, that is, the grain from which the subpericarpal layers had been polished off; (C) a quantity of the polishings, that is, the material removed subsequent to the separation of the husk and which includes the pericarp with the subpericarpal layers. The miller estimates that 40 parts of paddy produce 25 parts of white rice, 5 parts of polishings, and 10 parts of husk. The polishings are sold as food for cattle and the husks are burned as fuel in the mill.

Experiment A.—Twelve fowls were fed on the husked grain for five weeks.

Result: All remained healthy.

Experiment B.—Twelve fowls were fed on the white rice alone.

Result: In five weeks six had developed polyneuritis; two were dead, one having suffered from polyneuritis and one from a disease other than polyneuritis; five fowls remained healthy.

Experiment C.—Twelve fowls were fed on rice taken daily from the same bag as that used in experiment B; in addition, polishings in the form of emulsion, in amount equal to that milled from the quantity of rice consumed, were fed daily by a tube passed into the crop. This quantity was subsequently diminished week by week until only 3 grams of polishings per kilo of body weight were being given daily. This amount sufficed to maintain the fowls in health and in constant weight.

Result: The experiment was continued for seven weeks and all remained healthy.

The result was subsequently confirmed for rice taken from places where known outbreaks of beriberi had occurred.

It will be understood that these three experiments were in progress simultaneously and that the fowls were in all respects under identical conditions.

Experiment D.—Part of the original paddy was taken and milled by a Malay woman by primitive methods into the finished product as eaten by Malays. Eight fowls, fed for five weeks on the rice prepared from the original paddy by the Malay method, remained healthy. Eight fowls only were used for this experiment, as the quantity of paddy then remaining sufficed only for this number for the time it was estimated the experiment would last.

Attention is drawn to the important point that the products used in these experiments were all derived from the same lot of paddy, and the results force us to the conclusion that it is the polishing process, which is essentially at fault; the polishing of white rice removes from the seed some substance or substances essential for the maintenance of the normal metabolism of nerve tissues.

To elucidate the point as to whether rice when freshly milled is less harmful than that which has become stale, an assistant was stationed in Singapore who sent daily to the laboratory by the most expeditious route a quantity of rice milled on the day of dispatch. Twelve fowls were fed on this rice and five developed polyneuritis in four weeks. This result, which is similar to that obtained in other experiments, when fowls were fed on rices milled from four weeks to two years previously, disposes of the suggestion that the harmfulness of white rice is due to its staleness or the development in it of a poisonous substance or substances subsequently to its being milled. The root of the evil lies in the milling process itself. The result further indicates the inadequacy of preventive measures founded on the poison hypothesis in regard to the use of freshly milled rice.

An experiment was now planned to determine whether a parboiled rice proved harmless, could by exhaustion with hot alcohol be reduced to such a condition that it would produce polyneuritis when fed to fowls, and whether the substances so extracted when fed to fowls with a white rice proved harmful could prevent the development of polyneuritis. For this purpose parboiled rice was repeatedly exhausted with hot alcohol. The alcoholic extracts were concentrated *in vacuo* at a temperature of 52°, freed from alcohol and the residue emulsified in distilled water. Experiments with these products showed that fowls fed on the exhausted, parboiled rice contracted polyneuritis, and that birds fed on a white rice proved harmful by previous experiment remained healthy if they received in addition a quantity of the extract.

Having by these and other experiments, the details of which are omitted so as not to encumber the argument, arrived at the point where it was clear that the essential cause of beriberi was to be sought for in a nutritive defect, further efforts were made to determine by chemical

methods precise differences between various rices. Such differences, if they are to furnish an adequate explanation for the origin of beriberi, must be in accordance with clinical observations and the experimental results in fowls.

In view of the important rôle played by phosphorus compounds in the metabolism of nerve tissues, the amount of phosphorus in various kinds of rice was determined as phosphorus pentoxide. The result of a large series of observations showed that a reduction in the amount of phosphorus pentoxide obtained from rice was directly related to the probability of the rice producing beriberi; in other words, the higher the phosphorus content of a rice the less was the liability of that rice to produce the disease, and *vice versâ*.

Thus, a sample of parboiled rice which was fed to fowls over many weeks all remaining healthy, was found to contain 0.469 per cent P_2O_5 and a sample of white rice which produced polyneuritis in fowls yielded 0.277 per cent P_2O_5. The rice polishings employed in experiment C yielded 4.2 per cent P_2O_5.

From a series of observations it was determined that a fowl under the conditions of our experiments, weighing from 1,200 to 1,400 grams, required 60 grams of parboiled rice daily to maintain it in health and in nutritive equilibrium. In experiment C it was determined experimentally, the chemical analysis being then unknown, that when fed on white rice a fowl of this weight required the addition of about 3.5 grams of polishings to preserve it in nutritive equilibrium. From the data given above it may readily be calculated what amount of polishings added to white rice, is required to raise the phosphorus content of the white rice diet to that of the parboiled rice. Thus

	Grams P_2O_5.
60 grams of parboiled rice	0.3120
60 grams of white rice	0.1662
Difference	0.1458

Polishings contain 4.2 per cent of phosphorus pentoxide.

Calculated from the phosphorus content, therefore, 3.47 grams of polishings added to the 60 grams of white rice supplied to a fowl of 1,200 to 1,400 grams weight should preserve it in nutritive equilibrium. From experimental observation 3.5 grams of polishings had been shown to accomplish this result. This can scarcely be regarded merely as a coincidence, but its exact significance and importance can not yet be estimated.

Fowls receiving nothing but water do not develop polyneuritis, while fowls receiving only polished rice and water do. No satisfactory explanation of this observation has as yet been obtained, but further researches are in progress. Meanwhile the amount of phosphorus estimated as phosphorus pentoxide contained in a given rice may be used merely as an indicator of its liability or otherwise to produce beriberi.

We are greatly indebted to Mr. B. J. Eaton, chemist in this institute,

for valuable assistance in the chemical part of this investigation, and to Dr. R. D. Keith for suggestions as to methods for the examination of the nerves.

SUMMARY.

1. Beriberi is a disorder of metabolism and, as it occurs in this country, is associated with a diet in which white rice is the principal constituent.

2. White rice as produced in the mills here commonly makes default in respect of some substance or substances essential for the maintenance of the normal metabolism of nerve tissues. These substances exist in adequate amount in the original grain and in superabundant amount in the polishings from white rice.

3. The estimation in terms of phosphorus pentoxide of the total phosphorus present in a given rice may be used as an indicator of the beriberi-producing power of such rice when forming the staple of a diet in man.

The prevention of beriberi in this country will be achieved by substituting for the ordinary white rice a rice in which the polishing process has been omitted, or carried out to a minimal extent, or by the addition to a white-rice diet of articles rich in those substances in which such white rice now makes default. One such article which is cheap and readily obtained is the polishings from white rice.

The use of parboiled rice as suggested by Doctor Braddon will achieve a like result, provided that the polishing process is not carried beyond the limited extent now customary.

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ILLUSTRATIONS.

PLATE I.

FIG. 1. Fowl-run, showing arrangement of cages.
2. Fowl fed on white rice. Early stage of polyneuritis.

PLATE II.

FIG. 3. Fowl fed on white rice. Polyneuritis.
4. Fowl fed on white rice. Late stage of polyneuritis.

PLATE III.

FIG. 5. Teased preparation of sciatic nerve of fowl suffering from polyneuritis. Wallerian degeneration.
6. Cross section of rice grain, after removal of the palea or husks, showing the pericarp and subpericarpal layers intact.
7. Cross section of rice grain treated by parboiling before milling, showing the subpericarpal layers intact.
8. Cross section of grain of white rice (Siamese). Subpericarpal layers have been removed in polishing.

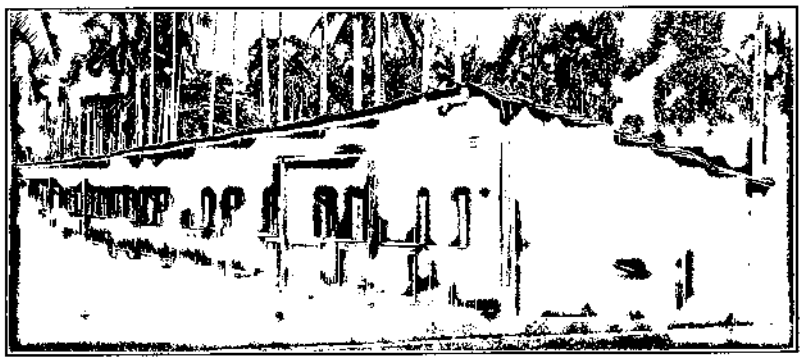


FIG. 1. FOWL RUN, SHOWING ARRANGEMENT OF CAGES.

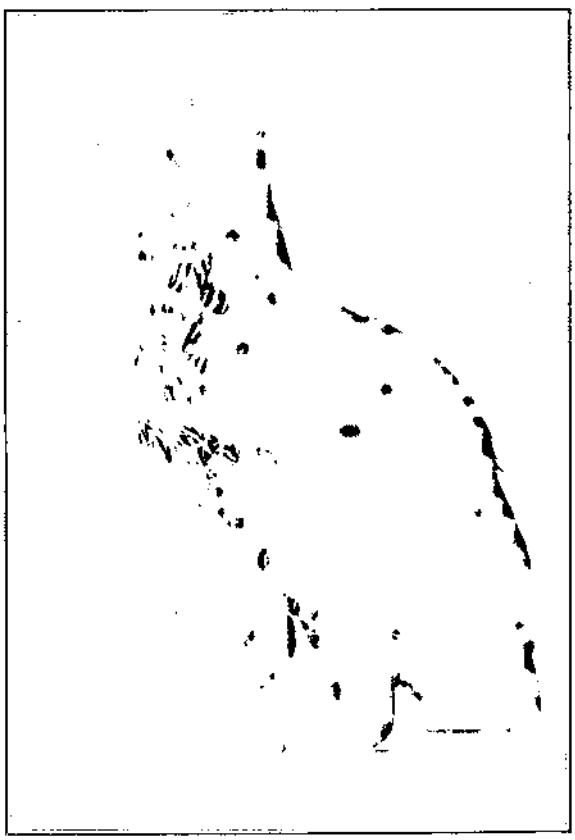


FIG. 2. FOWL FED ON WHITE RICE. EARLY STAGE OF POLYNEURITIS.

PLATE I.

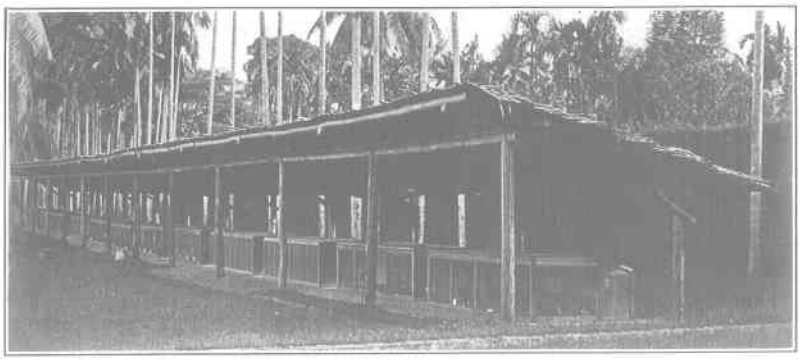
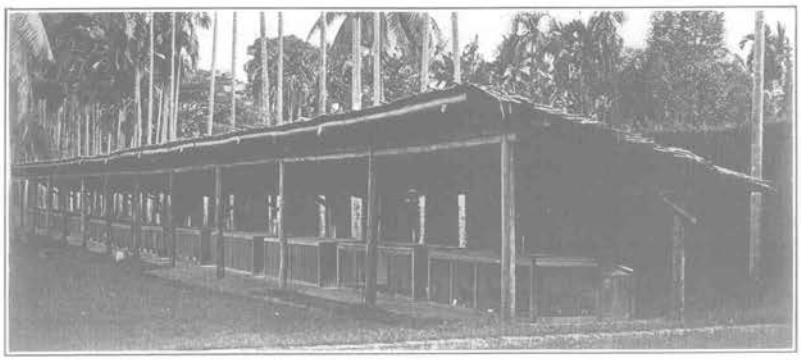


FIG. 1. FOWL RUN, SHOWING ARRANGEMENT OF CAGES.

FIG. 2. FOWL FED ON WHITE RICE. EARLY STAGE OF POLYNEURITIS.

PLATE 1.



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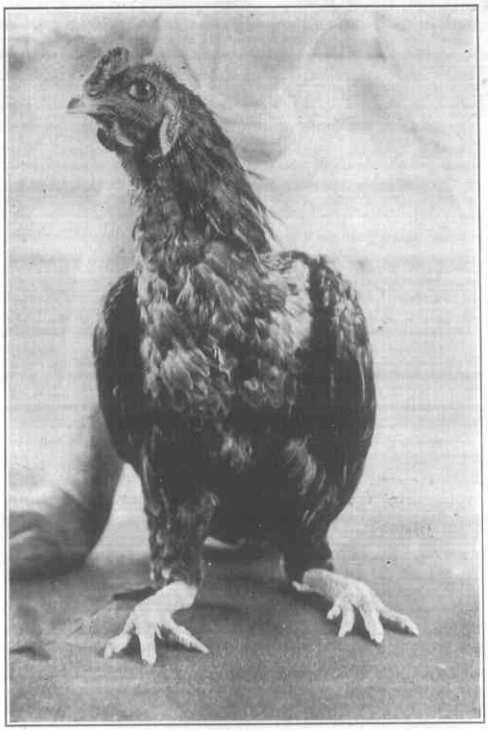

FIG. 3. FOWL FED ON WHITE RICE. POLYNEURITIS.

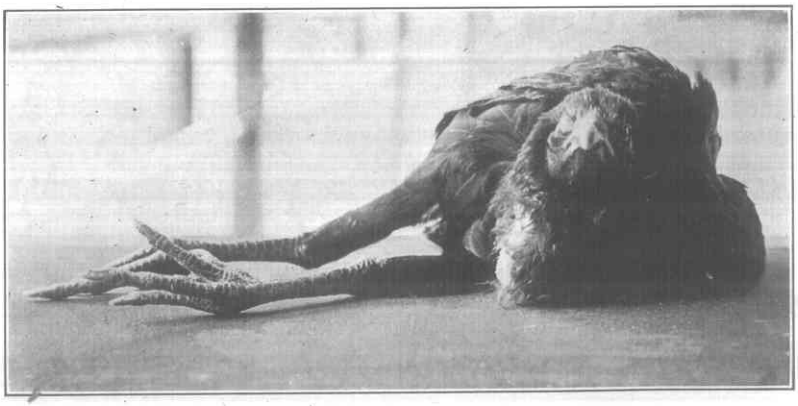

FIG. 4. FOWL FED ON WHITE RICE. LATE STAGE OF POLYNEURITIS.

PLATE II.

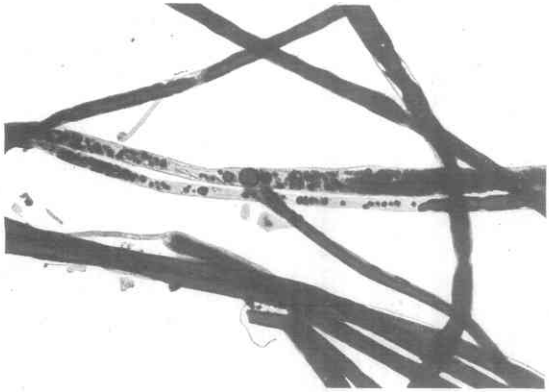

FIG. 5. TEASED PREPARATION OF SCIATIC NERVE OF FOWL SUFFERING FROM POLYNEURITIS. WALLERIAN DEGENERATION.

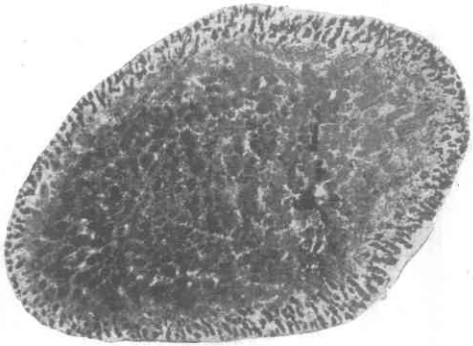

FIG. 7. CROSS SECTION OF RICE GRAIN TREATED BY PARBOILING BEFORE MILLING, SHOWING THE SUBPERICARPAL LAYERS INTACT.

FIG. 6. CROSS SECTION OF RICE GRAIN, AFTER REMOVAL OF THE PALEÆ OR HUSKS, SHOWING THE PERICARP AND SUBPERICARPAL LAYERS INTACT.

FIG. 8. CROSS SECTION OF GRAIN OF WHITE RICE (SIAMESE). SUBPERICARPAL LAYERS HAVE BEEN REMOVED IN POLISHING.

PLATE III.

ON THE ETIOLOGY OF BERIBERI.[1]

By J. DE HAAN.[2]

Beriberi has for many years been a subject of study by the government medical laboratory at Weltevreden, and the object of this paper is to communicate the results of all these investigations and to give the point of view which is maintained by us with reference to the etiology, especially for the Tropics, of this interesting disease.

No one hitherto has succeeded in proving beriberi to be caused by a specific microbe, or shown that it should be classed among the infectious diseases. Neither its epidemic nor endemic dissemination, nor the few cases mentioned in literature indicating the possibility of infection from one person to another, should be considered as a proof of its microbic origin, since the clinical symptoms of beriberi—polyneuritis with all its sequels—may be caused by many other factors. My own very numerous attempts to find the *causa morbi* in the blood, the organs, or the excreta of persons suffering from beriberi also have never succeeded, and although I should not wish to consider this as a decisive argument for a nonmicrobic infection, it must be granted that it increases its probability.

By degrees we have gathered a series of facts that may possibly throw some light on this obscure subject.

Doctor Eykman, in the year 1888, observed the appearance among the laboratory fowls of an epidemic of polyneuritis that in many respects resembled beriberi. The clinical symptoms were: Staggering, often followed by total paralysis, paresis of the wings, dyspnœa and cyanosis; followed by death. The post-mortem examination revealed emaciation of the subcutaneous fatty tissue as well as of the muscles, much fluid in the pericardium, degeneration of nerve fibers, but no macroscopic nor microscopic alterations of the brain or of the spinal cord. It was evident that this disease was not caused by an infection with microbes, but that

[1] Read at the first biennial meeting of the Far Eastern Association of Tropical Medicine, at Manila, March 11, 1910.

[2] Director of the Government Medical Laboratory at Weltevreden, Java, Dutch East Indies; delegate from Her Majesty's Government of the Netherlands East Indies.

it was closely associated with the food of the fowls. It always appeared after an incubation period of varying duration (ranging from twenty to thirty days, sometimes more) when the birds were fed on boiled rice, the same as was given to the hospital patients. The polyneuritis could not be caused by a poison present in the rice, for fowls sickened much sooner when fed on boiled rice than when the same variety of grain was given unboiled. When they were given unhusked rice, or the so-called red variety, no polyneuritis appeared; the sick ones could even be cured by the latter.

It then became of great importance to determine the difference between white and red rice. No distinction can be detected between these two varieties when they are unhusked, but it becomes apparent after they have been husked. The grain (endosperm) in the red rice had retained its pericarp (inner skin—Dutch, *zilvervlies*), because there is great coherence between the latter and the endosperm; the pericarp contains a red color in its component cells. Red rice is commonly used as an article of food for animals, and it is eaten by man only in some parts of Java, therefore, no great labor is spent on removing the pericarp. The pericarp must be totally removed to render the grain suitable for the table (cleaned, or white rice), after which manipulation the endosperm alone remains, that is, the grain then consists almost entirely of starch.

These experiments prove that the husks are of no importance in the etiology of polyneuritis. On the other hand, when the diet consists of rice alone, the appearance of the disease depends upon whether the pericarp has been removed or not. Sick fowls recovered when fed on cleaned rice, together with a quantity of the bran from the rice.

Fowls fed on starch (cakes made of sago meal, pearl tapioca, or the starch of the sugar palm tree [3]) also suffered from polyneuritis, but they soon recovered, exactly as fowls did which had fallen ill by being fed on cleaned rice alone, when subsequently given raw meat. The disease also appeared when the birds were fed on sago, tapioca or sugar palm starch, together with a small quantity of meat; on the other hand, those which secured potato-starch, or milk sugar with a small quantity of meat, remained healthy. Polyneuritis never appeared when the fowls were not especially fed. Pigeons contract the disease by being given the same diet as fowls, whereas mammals do not.

From the results of his experiments, Doctor Eykman concludes that polyneuritis is caused by a poison present in starch or developed from it, and that there are one or more constituents in the pericarp of rice which counteract the poison, or prevent its formation. The same conclusions hold with all varieties of rice; neither its origin nor the time during which it has been stored are of the slightest importance.

[3] *Arenga saccharifera* Labill.

Doctor Eykman's investigations, mentioned above, from 1888 to 1896 caused the sanitary inspector, Doctor Vorderman, to undertake a tour of inspection through all the prisons in Java and Madura to gather exact data on the prevalence of beriberi during the year 1895 and the first half of 1896, and to collect samples of rice in order to discover whether any connection could be established between the prevalence of beriberi in the different prisons and the kind of rice consumed therein.

The result of this inspection was as follows:

Out of 52 prisons, where cleaned rice constituted the principal article of food, 37 were affected with beriberi, and the average percentage was 71.15. Out of 37 prisons, where rice with the whole of the pericarp constituted the principal article of food, 1 contained beriberi cases, and the average percentage was 2.7. Out of 12 prisons, where half-cleaned rice constituted the chief article of diet, 5 showed beriberi to be present, and the average percentage was 41.46.

The conclusion deduced from these figures, namely, that among men a connection also exists between the prevalence of polyneuritis and rice eating, although criticised by some, must be maintained.

Last year I made a trip to the coal mines of Sawah Loents in Sumatra. Cases of beriberi among the coolies working in the mines at this place are very rare, and I ascertained that the rice eaten by these men was half cleaned.

Statistics on the frequency of beriberi among the Singkehs in the tin mines at Blinjoe in 1908 apparently prove that beriberi was encountered twenty-four times as frequently among the coolies who eat cleaned rice as among those who eat grain which has not lost all of its pericarp.

Doctor Grijns continued these investigations in our laboratory after Doctor Eykman's departure for Holland in 1906. He did not succeed in preventing an outbreak of polyneuritis among fowls fed on cleaned rice by adding to the latter the different salts or the fat, contained in the grain in a smaller quantity than in the pericarp. When the pericarp itself was added in large quantity, then polyneuritis did not appear; but small quantities of the pericarp only delayed its incidence. It was impossible to extract the active constituents from the pericarp.

The beans of *Phascolus radiatus* Linn., termed *katjang idjo* by the Javanese, not only proved to be capable of preventing an outbreak of the disease among fowls fed on boiled, cleaned rice, but even when it had developed, the birds could very easily be restored to health by eating *katjang idjo*. Another variety, *Cajanus indicus* Spreng. (*katjang iris*), gave the same result. Investigations made in 1909 proved that many other kinds of beans, namely, *katjang bogor* (*Voandzeia subterranea* Thou.), *katjang tjina* (*Arachis hypogæa* Linn.), *katjang pandjang* (*Vigna sinensis* (Linn.) Endl.,) (*V. catjang* (Linn.) Walp.) have the same properties.

Katjang idjo loses its prophylactic and curative power when steamed

at 120° for from one to two hours. Fowls even fell ill when fed on steamed beans alone, but it was possible to keep birds which were given steamed *katjang idjo* to which only a small quantity (a few grams) of raw beans had been added, in good health and even sick birds could be cured by the latter mixture. Ducks also contracted polyneuritis when fed on cleaned rice. Fowls also became ill when given unhusked rice, or meat steamed at 120° during two hours. Pigeons fed on meat extracted by boiling, died of polyneuritis.

The disease also appears among fowls fed on raw, unhusked rice, although these instances are very rare. Doctor Eykman, in his later experiments, could not confirm this observation made by Doctor Grijns, but I was able to do so. I kept birds in my own garden, where they were fed only on unhusked and red rice; among these was one cock which suffered repeatedly from the clinical symptoms of polyneuritis and each time was cured by *katjang idjo* and raw meat. The late Mr. de Bruin, of the Veterinary School at Utrecht, observed polyneuritis in a cock fed on a mixture of maize, buckwheat, barley, and husked rice. However, these observations can not be used as an argument against a connection between polyneuritis and rice eating, because polyneuritis may also be produced by many other factors.

Doctor Grijns, in contradistinction to Doctor Eykman, observed that fowls when given milk sugar and potato starch, together with steamed *katjang idjo*, contracted polyneuritis.

Doctor Eykman, after arriving in Holland, repeated the same experiments and, after some failures, produced results identical with those which he had secured in our laboratory. He proved barley, rye, oats, and millet to be excellent food for fowls, but they caused polyneuritis when steamed in an autoclave at 110° to 125°. Sick fowls could be cured by an aqueous extract of rice bran. Potato starch, which he found in his former investigations was incapable of causing polyneuritis, produced the disease when steamed for two hours at 125°. He could not confirm the result obtained by Doctor Grijns in producing polyneuritis by feeding steamed meat.[3] Doctor Eykman, owing to his new experiments, reached the same conclusions as he did from those of 1889 to 1896, namely, that an outbreak of the disease depends upon the presence of definite varieties of starch in the food. He again concluded that the degeneration of the nerve fibers is brought about by a poison which is developed from this starch during digestion. He assumes constituents capable of preventing the formation of this poison to be present in different varieties of food, but believes these to be destroyed by heating to 110° to 125°.

[3] A. Holst, Experimental studies relating to ship beriberi and scurvy." *Journ. Hyg.* (1907) 7, 619–671, stated that chickens fed on meat steamed at 120° during two hours died from polyneuritis.

Doctor Grijns, after he succeeded in producing polyneuritis in fowls as well as pigeons by feeding them on food without starch, concluded that the nervous system requires definite and until now unknown constituents which are present in different articles of food in widely varying quantities; *katjang idjo* containing a great proportion of the latter, cleaned rice only a few. These constituents can be rendered inactive by steaming at 120° and an article of food which only contains small amounts of them is capable of causing disease of the peripheral nervous system. This author has for a long time been occupied in separating the active constituents from *katjang idjo*, but has not as yet been successful.

Doctor Schaumann, as a result of his investigations on ship beriberi, believed it possible that the nucleins are indispensable to the nervous system. He came to this conclusion because in cases of beriberi the secretion of compounds of phosphoric acid is diminished, and articles of food which are considered as probable factors in the etiology of beriberi, such as, for example, cleaned rice and dried potatoes, contain but a small amount relatively of these compounds, whereas the pericarp of rice and *katjang idjo* have a great quantity. He also proved that a large proportion of the nucleins is decomposed in old beans, which is a reason why articles of food in good condition, by reason of a long voyage or by being stored for a long time, may become unfit to supply the nervous system with sufficient nutriment.

After this communication, Doctor Grijns attempted to cure fowls suffering from polyneuritis by means of nucleins derived from *katjang idjo*, but with negative results. Doctor Schaumann brought forward the same results in his later publications, for in the case of pigeons suffering from polyneuritis, he could produce no effect by giving nucleins prepared from yeast.

In 1909, Doctor Grijns published the following investigation:

He injected into the peritoneal cavity of healthy fowls the blood of diseased birds (a minimum of 57 cubic centimeters in 12 injections and a maximum of 220 cubic centimeters in 23), but without result. The nerves of diseased fowls, either free or inclosed in collodion sacks and introduced into the peritoneal cavity of healthy birds, did not give rise to polyneuritis. The feeding of healthy fowls on the flesh of diseased birds had no effect, neither did the injection of the blood of the former into the latter accelerate the appearance of polyneuritis in the case of fowls fed on cleaned rice.

In the year 1901 an epidemic of beriberi occurred among the coolies at the coaling station at Sabang (Sumatra). The medical officer there present, Dr. H. L. Roelfsema, could not observe any amelioration in the condition of the patients when he gave them meat and other extra articles of food, but he did observe that the epidemic ended as soon as he prescribed *katjang idjo*. Doctor Hulshoff-Pol repeated these experiments in the lunatic asylum at Buitenzorg. During the period from August 1

to April 30, 1902, the patients from twelve pavilions of the asylum received the following in addition to the ordinary diet:

In 3 pavilions, 150 grams of *katjang idjo;* in 3 pavilions, 300 grams of fresh greens; and in 6 pavilions, ordinary diet. The pavilions were disinfected once a week with carbolic soap, 3 per cent, in order to kill any insects which might be of importance in the dissemination of beriberi.

The following were the results: Seventy lunatics who ate *katjang idjo* did not develop any cases of beriberi. The 86 who were given fresh greens gave 16 cases, and 33 of the 78 who lived in the disinfected pavilions contracted the disease. There were 58 control patients, of whom 19 contracted beriberi.

The curative power of *katjang idjo* was proved by the following:

Out of 64 patients suffering from beriberi, 44 were treated with *katjang idjo,* and these recovered; 20 were left without these beans, and of these 7 died, 6 recovered or improved, and 7 became worse; but after the use of *katjang idjo* the latter also became well.

Doctor Hulshoff-Pol proved that a decoction of *katjang idjo* has the same curative and preventive power as the beans themselves, not only when given to patients suffering from beriberi, but also to fowls with polyneuritis. He prepared the decoction by boiling 1,000 grams of *katjang idjo* with 2.5 liters of water during one and one-half hours, until only 1 liter remained.

In 1908 Dr. Kiewiet de Jonge, in our laboratory, repeated Doctor Hulshoff-Pol's experiments on 384 patients in the lunatic asylum at Buitenzorg. *Katjang idjo* was given to 182 of these, but not to the remaining 202. The result was as follows:

(A) As to the curative action of *katjang idjo:*

Suffered from beriberi and—	With *katjang idjo,* per cent.	Without *katjang idjo,* per cent.
remained unchanged	15.0	23.4
improved	75.0	13.3
became worse	10.0	63.3
died	2.5	30.0

(B) As to the prophylactic action of *katjang idjo:*

Had no beriberi and	With *katjang idjo,* per cent.	Without *katjang idjo,* per cent.
did not contract the disease	97.2	76.2
contracted it	2.8	23.8
contracted it and died	0	9.3

Thirty-six patients were treated with a decoction of *katjang idjo.* The symptoms of the disease either were greatly ameliorated, or totally disappeared among all of them.

In the year 1909, together with Doctor Grijns, I published a series of experiments describing our attempts to prove the presence of antibodies in the blood serum of beriberi patients, or of fowls suffering from

polyneuritis. We did not succeed in proving them to be present. Neither was it possible to find antigen in any of the various organs. We desire to draw attention to this work as giving further confirmation of the identity in the character of the experimental polyneuritis of fowls and polyneuritis epidemica, or beriberi, in man.

The following conclusions are drawn from the above-mentioned investigations.

CONCLUSIONS.

1. The disease described as beriberi, or polyneuritis epidemica, bears a very great resemblance in its etiology, prophylaxis, and therapeutics to the polyneuritis which can be artificially produced in animals, principally in fowls.

2. Just as is the case in the latter, the former is almost always the result of eating cleaned rice, that is, rice deprived of the whole of its pericarp, although other articles of food, prepared in a special manner, may also cause an outbreak of the disease.

3. The pericarp of the rice and also some parts of the grain are removed in the manipulation, by which unhusked rice becomes cleaned rice. Certain constituents of the greatest importance in securing normal nutriment of the peripheral nervous system are lost during this operation.

4. The constituents are neither salts, nor nucleins.

5. Their character is still unknown.

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BERIBERI IN SIAM.[1]

By H. Campbell Highet.[2]

Beriberi as a cause of invaliding and of death is now a common enough disease in Bangkok, but it has not always been so; in fact, the only reliable records which I can obtain date no further back than the year 1890. In that year an epidemic broke out in the central jail and, in the absence of definite figures, I can not do better than to quote a portion of a letter to me with general reference to beriberi, by Dr. Heyward Hays, of Bangkok. He writes:

My first personal contact with the disease occurred in 1890 in the new jail. Doctor Willis, who was then the physician to the British legation, invited me to go to the jail with him to see a number of cases he supposed to be beriberi. I confirmed his diagnosis. It was of the wet variety and very fatal. Doctor Willis and myself made out a report which was handed to His Siamese Majesty and the recommendations made in that report were immediately granted and carried out. The result was that the disease disappeared and was not seen in the jail up to the time I resigned in 1898, having succeeded Doctor Willis as physician to the jail in 1892. My next experience with the disease was in the year 1896 at Chantook and Muet Lek, during the construction of the Korat Railway.[3] We had a great many cases, particularly of the wet variety, which was very fatal. My next experience with the disease was in the year 1897 at Java, during His Majesty's visit there. It broke out on all our ships simultaneously and we had some eighty or ninety cases. None proved fatal, owing to the fact, I believe, that the disease was discovered in its early stages, as well as to the caustic measures which were taken to stamp it out.

Beyond these epidemics of the disease reported by Doctor Hays, beriberi was unknown to the general practitioner and even to the hospital physician until 1900. Arriving in Bangkok in April, 1897, after nearly five years' practice in Singapore, where I had seen much of the disease, I was soon struck with the total absence of cases of beriberi amongst my patients, whether in hospital or in private practice. On inquiry of

[1] Read at the first biennial meeting of the Far Eastern Association of Tropical Medicine, March 11, 1910.

[2] Fellow of the Royal Institute of Public Health; principal medical officer, local government, Bangkok, Siam; delegate from His Imperial Siamese Majesty's Government.

[3] These places are over 60 miles from Bangkok.

several of my colleagues who had already been many years in Siam, I was told that beriberi was a rarity. However, on August 29, 1900, our first case was admitted to the Police Hospital in the person of a Siamese constable. Another case came in a month later, but after that no further ones are noted in our Police Hospital register until April, 1901, since which time beriberi has become one of the ordinary diseases seen in that hospital. During this year, 1901, a very severe epidemic broke out amongst the soldiers in the central barracks, several cases were reported from the navy, and cases began to be admitted in increasing numbers to the general hospitals.

Since then, beriberi has taken a prominent place as a cause of invaliding in the public services. In the absence of reliable data for the ordinary civilian population, the following return of cases of and deaths from beriberi recorded in the hospitals, the army, the navy, and the police during eight years may be of interest:

Year (April to March).	Cases.	Deaths.
1901–2	1,128	14
1902–3	1,007	81
1903–4	2,615	161
1904–5	2,813	108
1905–6	3,861	92
1906–7	2,712	101
1907–8	2,427	229
1908–9	4,607	282
Totals	22,670	1,063

It must be noted, however, that these 1,063 deaths give no real conception of the actual death rate, as it was the custom of us all to send our beriberi sick away to their own homes in the country, finding that after a month or two many returned cured. Why such cases should recover much more satisfactorily if sent home is a most important point which will be discussed later. At any rate, here we have a total of nearly 23,000 cases in the public services in eight years. Naturally, the figures for the present year are not yet obtainable, but I have no doubt they will be very high. Compulsory registration of deaths only came into force in Bangkok in October last, but during the three months during which the law has been in force 203 deaths from beriberi have been recorded. So much for the history of beriberi in Bangkok up to a year ago.

Now, as to the cause of beriberi. My experience in Bangkok leads me to confirm absolutely the opinion first expressed by Braddon and lately confirmed by Fraser and Stanton, of Kuala Lumpur, that beriberi is closely associated with the consumption of white, steam-milled rice.

Why this should be so, I am not yet prepared to say. Is it due to the presence of a fungus on the white rice, which has become stale, is it

the result of want of nitrogen, or is it due to the removal of the oil-containing rind of the grain and so leading to fat starvation? These are points which are still *sub judice* and we await with much interest the elucidation of this fascinating problem. From a practical administrative point of view, at least, we may definitely say that beriberi is apt to attack those who regularly consume white, steam-milled rice, and in support of this view I shall now give you the facts from Bangkok.

1. Beriberi was, so far as we can find out, unknown in Bangkok until white, steam-milled rice began to be retailed locally. It is true that for over ten years before the first outbreak in the jail, white rice was prepared in one or two steam mills in Bangkok, but the whole of this was exported to Europe. One of the first large institutions to be supplied with white, steam-milled rice was the new jail, with the result already recorded. Doctor Hays tells me that it was the prisoners themselves who suggested a return to hand-milled rice, which they had always been accustomed to eat. The change having been made, beriberi died down and has since then given practically no trouble, as the prison authorities have continued to supply this form of rice, which is actually prepared in the jail by convict labor.

During the nineties, attracted by the immense profits which were being made by owners of steam rice mills, many mills were erected, and soon large quantities of white rice were thrown upon the local market. Two factors played into the hands of the rice millers. The first was the abolition of slavery in Siam. Hitherto, the preparation of the household rice was carried on by slave labor, but on the abolition of this practice the price of labor naturally went up. The second factor is a natural corollary of the former, for steam-milled rice could be produced at a much cheaper rate than that manufactured by paid hand labor. By the year 1900 most of the old hand mills had stopped working, and on a diet of steam-milled rice the people began to suffer from beriberi; our first cases being admitted into the hospital during this year.

2. A second fact in support of the white rice theory is the history of our experience in the asylum for the insane. In the year 1900 steam-milled rice was substituted for hand-milled, and beriberi soon appeared. During nine years dieting upon this rice the disease steadily increased in virulence and, in all, 763 patients died of the disease. Early in February, 1908, I determined to try the effects of parboiled rice. Hand mills and all the other apparatus necessary for the purpose were installed in the asylum. We purchased our own paddy, parboiled it, milled it by hand and issued it for the first time on February 15.

Since then not a new case of beriberi has developed amongst those patients who have continued to eat this form of rice, and it is now a year since it was first issued. No other alteration in diet or in hygienic surroundings was made in the asylum.

3. Still another fact in favor of the same theory comes from the Reformatory School at Koh Si Chang in the Gulf of Siam. I can not do better than to quote a note upon this subject forwarded to me by Mr. E. St. J. Lawson, the commissioner of police under whose charge the reformatory is placed. The "station" mentioned is the police station on the island. Mr. Lawson writes:

> The reformatory was started on March 5, 1908. There were no cases of beriberi either amongst the police or in the reformatory until I sent steam-milled rice from Bangkok. I did this because the men complained of the high price of rice on the island and asked me to buy and send to them. In the middle of February, 1909, I started sending hand-ground rice for the station and school use, and changed this to parboiled rice in April. There has not been a single case of beriberi, either in the reformatory or station since the change from steam-milled rice.

To supplement Mr. Lawson's remarks, I may say that there was no medical man on the island, that only serious cases were sent up to Bangkok, that 5 of these, out of a total of 50 boys, were admitted to the hospiatl between November, 1908, and the middle of February, 1909, and that I can not state how many mild cases there may have been.

However, a change of rice from steam-milled to hand-milled and then to parboiled rice, as soon as we could supply it from the asylum, prevented any further cases.

4. The jails in the outlying districts of Bangkok afford us a further proof. In four of these jails, fresh, hand-milled rice is supplied to the prisoners, and in these beriberi is a distinct rarity. However, in two of these jails, steam-milled rice is supplied and beriberi is frequent. Unfortunately, I have not yet obtained details with regard to prisoners either admitted suffering from beriberi, or developing the disease in these jails, or as to the daily average number of prisoners under observation, but the rough figures for the year 1908–9 are as follows: Four jails on hand-milled rice give 5 cases and no deaths; 2 jails on steam-milled rice give 20 cases and 8 deaths.

5. From an administrative point of view, my most striking experience was gained at the Police School in Bangkok. Conscription for the Bangkok police having come into force, some 400 conscripts were admitted to the Police School early in January, 1909. The rice was white, steam-milled, and was supplied by a contractor. It was frequently inspected along with the other articles of food, and on all occasions appeared to be of good quality. However, within a fortnight after commencing to eat this rice, beriberi broke out and at the end of a month 353 out of a total of 400 conscripts had contracted the disease. Practically all these men had come from the country districts outside of Bangkok and most of them had been accustomed to eat only fresh, hand-milled rice.

All the sick men were sent home on a month's leave, at the end of which time very many returned well, many were much improved, while

a few were still far from well. On April 31 a second batch of conscripts was admitted, numbering again about 400. By this time, hand mills had been erected at the school and apparatus for the preparation of parboiled rice installed. The healthy were given fresh, hand-milled rice, the sufferers from beriberi parboiled rice. Since then, nearly a year ago, over a thousand men have passed through the school and only a few cases (14 in all) have been detected; and it is more than probable that in the latter instances the men came in already suffering from beriberi. The police prisoners have been fed entirely on parboiled rice and throughout the past year not one case has developed.

6. The last evidence which I shall bring forward in support of the white, steam-milled rice theory is the apparent geographical distribution of the disease in Siam.

In the Province of Bangkok, some 1,700 square miles in extent, we find that beriberi clings to the banks of the Menam River and to the banks of the large, navigable canals which join this river with the adjacent streams throughout the flat, alluvial plains in the neighborhood of the capital. Why is this? Because the river and these canals are the principal means of transport, and along these steam-milled rice from Bangkok is freely hawked. Back from the banks, where communication is difficult, we find that the cultivators mill their own rice and by so doing invariably escape beriberi. Further, amongst a total of 4,550 *gendarmes* scattered throughout the interior of Siam and fed entirely upon hand-milled rice only 6 cases of the disease were reported during the year 1908 to 1909. These 6 cases all occurred in one district, Nakorn Sawan, which stretches on both sides of the River Menam and to which steam-milled rice can gain easy access by boat.

To sum up these facts with regard to etiology: In Siam, as elsewhere in the East, the consumption of white, steam-milled rice would appear to be the principal factor, and the substitution of parboiled rice or of fresh, hand-milled rice is, so far as we know at present, the best practical method of preventing the disease.

Where there is difficulty in getting either of these forms of rice already prepared, see that good paddy is obtained and that it is milled fresh by hand every day. This is especially an important point in connection with coolies engaged on large engineering works, such as railways. In place of transporting white, steam-milled rice to the coolies, see that the food contractors either purchase paddy locally or transport it to the coolie lines, where native hand-mills should be erected and the paddy milled daily by hand. In large cities, where either parboiled rice or hand-milled rice is not obtainable in large quantities, try to induce steam rice-millers to prepare undermilled rice for local consumption.

White rice is the result of "overscouring," as the millers say, but I am told that rice similar to hand-milled can be supplied just as easily,

provided there is a demand for it. What is the essential difference between these various forms of rice? They may be divided into two classes, "overmilled" and "undermilled," the distinction being based upon the presence or absence of the pericarp.

White, steam-milled rice is so "overscoured" that none of the pericarp remains. It is overmilled. On the other hand, we find that hand-milled, parboiled, and machine-made "undermilled" rice all retain a considerable proportion of the pericarp, which gives a reddish tinge to these forms of rice in bulk. All three may be classed together as "undermilled."

The whole idea of prevention, then, would seem to be that rice, however milled, which retains a considerable proportion of the pericarp, does not cause beriberi, but that the removal of the outer layers of the grain by reason of the scouring process, either takes away a prophylactic agent, or renders the grain liable to deterioration and the consequent production of some active poison.

POSTSCRIPTUM.

Since writing these notes, prepared in haste on the eve of my departure for Manila, two important events, bearing upon my subject, have occurred. One of these was the receipt, while still in Bangkok, of the latest report by Doctors Fraser and Stanton on "The Etiology of Beriberi." On my voyage here to Manila I have carefully studied this very valuable contribution to the literature on beriberi, and, naturally, it is a matter of considerable satisfaction to me to find that the conclusions of these two able investigators so absolutely corroborate the results of our work in Siam. As they remark, "the root of the evil lies in the milling process itself; the polishing of white rice removes from the seed some substance or substances essential to the maintenance of the normal nutrition of nerve tissues." The deductions of Doctors Fraser and Stanton are the result of carefully conducted scientific experiments, assisted by chemical analyses; my experiments were practical administrative acts, forced upon me by local circumstances, but guided by observation of local conditions. The Siamese in the public services declined to eat parboiled rice owing to its stale, musty flavor. A substitute had to be provided in place of white rice, and this was found in the hand-milled grain.

With regard to parboiled rice, which is now so largely used in public institutions in the Straits Settlements and Federated Malay States, one point is, to my mind, clearly proved by Fraser and Stanton, namely, that its prophylactic powers are not due to any sterilization during the process of parboiling, but to the retention of a considerable proportion of the oil-bearing layers of the seed. This prophylactic property will be found to be distinctly more powerful if the parboiled paddy has been milled by hand.

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As I am now treating of the practical prevention of beriberi, and as time does not permit me to deal with any more of the many interesting facts brought forward by Fraser and Stanton, I shall only refer to one more point, namely, the suggestion that rice polishings be added to the diet of those using white rice.

Scientifically, this is a natural deduction from their experiments, but surely Fraser and Stanton must see that their advice is not practical. With people living in close proximity to mills in which white rice is prepared, it might be possible to obtain a supply of these polishings, but even then, in the case of the ordinary native, how can we expect that they will be able to gauge the proportion of the polishings required to make up for the deficiency in the white rice? Further, if the consumer lives in a country where rice is imported, I fear that the supply of these polishings would be a matter of difficulty. I would go even further and say that unless very carefully prepared, rice polishings would not be quite an ideal addition to a dietary, containing as they do much mineral débris from the grinders, dust, filth, etc. No; so far as I can see, the only practical method is to encourage the consumption of undermilled rice. How far legislation might be invoked in this question is too large a subject to be taken up now.

A second important fact learned on my arrival in Manila, is the abandonment by the American Army of the use of imported white rice and the substitution of locally grown and locally milled paddy.

Therefore, it would appear that we all have come to similar conclusions.

In conclusion, I would draw your attention to the name "Siam rice," so often used in writings upon the etiology of beriberi. This is a trade name, and amongst Siam rice we find grain from many of the other neighboring rice-growing countries. It is polished white rice of a certain quality which is inferred by this trade name. Naturally, as delegate from Siam, I am glad not only to have the opportunity of protesting against this use of the term in describing the cause of beriberi, but I believe that I have satisfactorily demonstrated the fact that Siam rice can be shorn of its dangers as a staple article of food through the simple process of "undermilling."

PHOSPHORUS STARVATION WITH SPECIAL REFERENCE TO BERIBERI: I.[1]

By HANS ARON.

(From the Physiological Laboratory, Philippine Medical School.)

It has been argued for a long time that there is a close connection between the food supply and the occurrence of certain diseases appearing in a more or less epidemic form, such as scurvy, beriberi and pellagra. Beriberi, as it appears in the Orient, has been the subject of especially careful studies, and the information which we have received as a result of these investigations allows us to make certain definite statements.

In spite of the claims of various investigators who have described a number of so-called beriberi organisms which all, more or less, have been proved not to be the specific cause of the disease, we can to-day regard it as proved that beriberi is *not an infectious disease*. Of course certain conditions of a general insanitary character, such as living in small, badly ventilated rooms, together with humidity and uncleanliness as well as the influence of tropical climate and other factors of environment, doubtless have some bearing upon the outbreak or occurrence of beriberi. However, the great successes which are recorded in limiting this disease by changes in diet prove beyond any doubt, when taken in connection with careful experimental investigations with different diets on people otherwise living under more or less similar sanitary conditions, that we must regard the diet as the main factor in causing beriberi.

It has been known for a long time in the tropical Orient that people living almost wholly or entirely on rice are more liable to contract the disease than are others. Eykman [2] has already proved that it is not rice in general which must be regarded as the cause of beriberi, but that certain kinds of rice, or, better, rice prepared in a certain way, are most liable to produce the disease. These observations, especially in late years, have

[1] Read at the first biennial meeting of the Far Eastern Association of Tropical Medicine, at Manila, March 11, 1910.

[2] Die Bekämpfung der Beriberi. *Virchow's Arch.* (1897), **149**, 187–194; *Ref. Maly's Jahresb. d. Tierchemie* (1897), **27**, 792.

been thoroughly confirmed by a number of authors.[3] All observers further agree on this point, that the same kind of rice which causes beriberi, if the diet of the people is based almost entirely on this foodstuff, is eaten without any ill effects if a sufficient amount of other things such as fresh meat, fresh vegetables, and especially certain kinds of beans are taken with it. An ample supply of fresh meat or of the beans, accompanied by a restriction of the quantity of rice consumed, or with a total change of diet, has always proved the best cure for beriberi.[4] The most striking fact in all these observations is the great influence which the preparation of the rice has in its relation to the etiology of beriberi. I will, in order to give a clearer understanding of this phase of the question, give a short description of the preparation of rice for consumption.

At first the rice husk must be removed from the grain (in Spanish and native dialect, *palay*). This husk (*ipa*) is never eaten. The grain (*pinaoa*) as it now appears when the husk is broken and thrown away is surrounded by a second, thinner skin of a more or less red or brownish color, which does not render the appearance of the rice very appetizing. This second skin can also be cleaned off by a process of milling between rotating stones which removes it, together with the outside layer of the rice grain; and after this process of milling or polishing, the grain is white and clean and now has the form which the European usually knows as rice. The first rice, which still contains the second skin (the *Silberhäutchen* of the German writers) is that which usually does not cause beriberi; while the grain which is deprived of this skin by the process of polishing must be regarded as quite liable to produce the disease. There are different degrees of polishing, for we find on the market rice which is only slightly polished and grain which has been very highly treated. There are, in addition, certain processes, usually practiced in India and elsewhere ("parboiling") which seem to prevent the thorough removal of the outer parts of the rice, but I am unable to judge of them, since they are not in use in these Islands. The native rice prepared by hand is pounded in a large mortar (Tagalog, *lusong*), and even if milled by a hand mill is never deprived of its outer layers to as great an extent as that milled by large machinery.

Before the exact origin and method of preparing rice capable of causing beriberi was known, it was termed "uncured," while that which did not produce the disease was called "cured" rice. However, while these names are very extensively used, I scarcely think that they convey the right idea.

[3] *Arch. f. Hyg.* (1906), **58**, 150–170; Fletcher, William. *The Lancet*, London (1907), **1**, 1776; Idem, *Journ. Trop. Med. & Hyg.* (1909), **12**, 127–135; Braddon, W. L. The Cause and Prevention of Beriberi. London. 1907. Idem. *Bombay Med. Congr.* (1909). Fraser, H., and Stanton, A. T. *Bombay Med. Congr.* (1909); An Inquiry Concerning the Etiology of Beriberi. *Studies from the Inst. for Med. Research. Kuhua Lampur* (1909).

[4] Walter, F. H. *Geneesk. Tyds. v. Ned. Ind.* (1908), **48**. Hulshoff-Pol, F. *Geneesk. Tyds. v. Ned. Ind.* (1909), **49**. (*Arch. f. Schiffs-u. Trop.-Hyg.* (1909), **13**, 775); Kiewit de Jonge, *ibid.*

I will, therefore, in conformity with Fletcher, designate the first rice as "unpolished" and the second as "polished," or apply the local names, "red" and "white" rice. The unpolished or red rice should be harmless, and we must regard the second process of polishing and milling as that which changes a harmless foodstuff to one harmful under certain circumstances.

This does not mean that the eating of "white" rice under all circumstances causes beriberi. In the first place, it is absolutely necessary that the grain should form the main constituent of the diet, and as the amount of other foodstuffs eaten is smaller in proportion to the quantity of rice consumed, the incidence of the disease under such circumstances is doubtless greater than it would be were a larger amount of other foodstuffs eaten. Furthermore, according to certain authors it seems that white rice stored for a long time, especially in humid rooms and similar places, favors the development of the disease more than the freshly pounded grain, but we must not forget that the latter is almost always prepared by the natives by hand and hence is not so highly polished an article as that kept in storage, which is prepared by machinery.

Eykman,[5] in the course of his investigations on beriberi has shown that chickens fed only on white rice develop a disease which in several respects is similar to beriberi. The most prominent symptoms are a paralysis of the legs and wings and an increasing weakness of the animal, which, if no steps are taken to prevent the further advance of the disease, finally dies. The pathologic examination shows a degeneration of the peripheral nerves, so that the condition has been termed *Polyneuritis gallinarum*. It is very interesting to note from our own standpoint that the same changes in diet, which either avoid or cure beriberi act in an absolutely similar manner upon the fowl.

The disease is only contracted if the fowls are fed on white rice. Those fed on the red grain will suffer very rarely, if at all. An addition of beans or fresh meat to the diet cures the chickens while in the early stages of the disease. Finally, a fowl fed on a sufficiency of beans and white rice or enough meat and white rice will not become ill at all. These experiments have been repeated by Grijns,[6] Maurer,[7] Holst and Fröhlich[8] and also recently by Schaumann,[9] and the observations of Eykman have been fully confirmed. These authors have shown that a similar disease can also be produced in pigeons. In another interesting series of experiments Eykman, Grijns and Axel Holst have further proved that not only white rice, but also certain other foods are liable to produce polyneuritis in chickens and pigeons, and that, especially, long sterilization at a high temperature of certain foodstuffs (such as horse meat) which are absolutely

[5] *Virchow's Archiv.* (1897), **148**, 523–532; *Arch. Hyg.* **58**, 150–170.

[6] *Geneesk. Tyds. v. Ned. Ind.* (1908), **48**; (1909) **49**.

[7] *Arch. f. Schiffs-u. Trop.-Hyg.* (1909), **13**, 233–252, 284–297.

[8] *Norsk. Magaz. f. Lægevidenskab.* **68**. (*Biochem. Centralb.* (1907), **6**, No. 1225; *ibid*, No. 2478.

[9] *Arch. f. Schiffs-u. Trop.-Hyg. Beiheft* V. (1908), **12**, 37.

harmless otherwise seems to favor the development of a polyneuritic disease. Holst and Fröhlich[10] were also able to demonstrate that guinea pigs fed on certain one-sided diets, consisting of various kinds of grain, groats, or bread, develop a disease in many respects similar to scurvy.

DIFFERENCES IN COMPOSITION BETWEEN VARIOUS KINDS OF RICE AND THEIR RELATION TO BERIBERI.

Why does the one variety of rice cause such severe sickness, whereas the other is eaten without any ill effects, and why does the identical, harmful rice not produce the disease if eaten in smaller quantities together with a sufficient amount of certain other foodstuffs?

This question aroused my interest in the study of this disease from the point of view of the physiology of nutrition. We know that fatal diseases can be caused by "spoiled" or bad foods, but these intoxications are brought about by certain *poisonous substances* formed in the food itself. We know further that certain artificially prepared articles of diet are unable to sustain life, but up to the present we have scarcely known any disease which is caused by an apparently normal foodstuff.

Numerous theories to explain the action of different classes of rice exist. In the first instance, a number of authors believe that certain fungi will grow better on the white rice than on the red, because the latter is protected by the pericarp, the introduction of these organisms, fungi, etc., being the real cause of the disease and the rice only an indirect factor. Others, and especially Eykman, regard the white rice as "poisonous," the antidote for all this unknown poison being present in the rice bran as well as in other antiberiberica. Maurer believed beriberi to be produced by the formation of oxalic acid from carbohydrates by a process of fermentation. These hypotheses directly contradict the exact results of experiment.

From a physiologic standpoint, the most probable explanation would be the assumption that by the preparation (milling the rice, sterilizing the food, etc.) certain constituents of importance are taken away, decomposed, or, at least changed so that they now no longer serve their purposes.

Eykman already has considered the subject of the chemical constituents taken away by the process of grinding and which therefore may be lacking in the white rice. If we compare the composition of polished and unpolished rice, we see that the grain by the process of milling becomes poorer especially in ash constituents, fat and cellulose, and also somewhat in protein.

[10] *Journ. Hyg.* (1907), 7, 619–672.

Balland[11] has shown this very plainly by a number of analyses which are given in the the following table:

Analyses of hand husked, machine husked, ground and polished rice.

Kind of rice.	Water.	Nitrogenous substance.	Fats.	Carbohydrates.	Cellulose.	Ash.
Husked by hand	11.00	9.05	2.80	64.98	1.12	1.10
Husked by machine	13.00	7.82	0.60	77.74	0.28	0.56
Husked and ground	12.90	7.82	0.40	78.20	0.24	0.44
Husked, ground and polished	13.30	7.65	0.30	78.18	0.21	0.36

The main constituents of the ash are the phosphates (53.7 per cent P_2O_5). Therefore Eykman has studied the question as to the extent to which this deprivation of phosphorus may cause the harmful action of white rice, but he could not find sufficient evidence to reach a definite conclusion. Two years ago Schaumann[12] endeavored to show that the disease termed "scurvy of sailing vessels," is caused by a lack of nucleophosphoric acid, which is extracted or destroyed by certain sterilization processes, and he argued that a similar lack of organic phosphorus (nucleoproteins) in the food, especially in the white rice, may produce beriberi.

It can easily be believed that a constant lack of phosphorus in the food may bring about a degeneration of exactly those tissues which are rich in phosphorus, namely, of the nerves. I took up the question myself at this point, directing my investigations especially toward the question of the influence of the phosphorus. Meanwhile, there has appeared a second publication by Schaumann,[13] continuing his researches, especially with experiments on animals, and quite recently, in a short paper Fraser[14] and Stanton stated that the phosphorus content of a rice is an indicator of its capability of producing beriberi.

The variations in the content of phosphorus are the most striking changes produced by the process of milling, as the following analyses of different classes of Philippine rice, made by me, will show:

Variety of rice.	P_2O_5.	Protein.
	Per cent.	*Per cent.*
Laguna rice, unpolished	0.557	9.00
Laguna rice, polished	0.314	7.87
"Macan," Bulacan Province, machinery rice	0.340	.....
"Macan," Bulacan Province, native made, freshly husked rice	0.455	.....
"Valenciana" rice, highly polished	0.197	.....
Average Manila rice	0.33	.....

[11] *Compt. rend. Acad. sci.* (1895), **121**, 561–566.

[12] *Loc. cit. Arch. f. Schiffs-u. Trop.-Hyg. Beiheft.* (1908), **12**, 37.

[13] *Arch f. Schiffs-u. Trop.-Hyg. Beiheft.* (1909), **13**, 82–90.

[14] The Etiology of Beriberi. *Studies from the Institute for Medical Research, F. M. S. Kulua Lampur* (1910).

I have further obtained a very striking proof of the fact that the rice with a low content of phosphorus will actually cause beriberi, whereas that with a considerably higher content will not, by the study of an outbreak of beriberi on an English steamer which came to this port. Through the courtesy of the captain of this ship I obtained the following data:

The Indian crew of the steamship *Knight Templar*, used rice almost entirely for food, because the religious laws of this people do not allow them to eat meat except that killed in a special way. For this reason, in addition to rice, only small amounts of mutton could be given to the crew not oftener than once a week. The identical crew had often before sailed on this vessel, and the captain, because of his own previous experience, wished to obtain a certain kind of Indian rice, which he always purchased at Calcutta and which he knew would in all probability not cause beriberi. This I will term "Calcutta" rice. It appears to be a grain which retains a great deal of the pericarp; it therefore is unpolished. The vessel on this particular voyage sailed from Bombay June 5, 1909. Another variety of rice was purchased in Bombay, which I will designate as "Bombay" rice. It was used as food for the crew and they showed no untoward symptoms.

The supply of rice taken from Bombay was exhausted when the vessel reached Liverpool, therefore a fresh supply was purchased. Unpolished rice, in spite of great endeavor, could not be purchased in Liverpool; the grain bought in the latter port ("Liverpool" rice) was of good appearance, white and highly polished. This was given to the crew, beginning about July 25, 1909, under the same conditions and in all likelihood in the same quantity as the "Bombay" rice on the voyage before.

On September 25 a member of the crew became ill and unable to work, but the sickness was not recognized by the captain; but when on October 1 and 2 a man on each day felt weak and could neither walk nor work, the captain then recognized the disease as beriberi. On October 16 the ship arrived at the port of Cavite (in Manila Bay). Here, upon the advice of the boarding officer, the two sick sailors were admitted to the United States Naval Hospital at Cañacao. After October 18 the disease increased so rapidly among the crew that 10 additional men were removed to the hospital on October 25. Here, through the courtesy of Dr. E. R. Stitt, United States Navy, I was able to examine the patients and to confirm the diagnosis of beriberi. One man in the hospital died (probably from paralysis of the diaphragm), but an autopsy was refused by the patient's friends.

The 11 men in the hospital were no longer given rice, but only European food, such as milk, eggs, meat, etc. They showed considerable improvement in a short time. "Bombay" rice and also Philippine red rice was fed to the remaining healthy crew, and only one light case of beriberi developed afterwards. The ship sailed on November 5 with the same crew, the latter having recovered to an astonishing degree in the short time which had elapsed after changing the food.

I have analyzed both varieties of the rice, samples of which I obtained from the captain, and also samples of his former "Calcutta" rice. The differences in the phosphorus content are exceedingly striking; slight variations in the nitrogen content are also to be noted. The following is the table of analyses:

Analyses of "Calcutta," "Bombay," and "Liverpool" rice.

Rice.	P_2O_5.	Protein.	Water.
	Per cent.	*Per cent.*	*Per cent.*
"Calcutta"	0.446	7.75	11.68
"Bombay"	0.408	7.94	11.97
"Liverpool"	0.148	6.69	11.67

Another occasion to study the connection between food and beriberi arose owing to an outbreak of the disease among the lepers at Culion. Here a careful, exact investigation of the data, such as was made in relation to the ship's crew, was impossible, because the people, or at least a part of the people at the leper colony, obtain their own foodstuffs, growing a little rice and catching a few fish, etc., but from the list of foodstuffs which I have given below, it will be seen that in Culion also, rice was the main constituent of the food. Beriberi, to a slight extent, practically was present at all times among the inmates of the colony. In November, 1909, the number of beriberi cases increased rapidly, with a still greater rise in December, 1909, and the first part of January, 1910. The following is the dietary:

Foodstuffs given at Culion per individual per day, in grams.

	August.	September.	October.	November.	December.	January.
Rice, about	700	700	700	700	600	600
Mongo[a]	8	25	36	15	28	90
Meat	40	30	35	40	35	35
Vegetables		26	30	50	35	25
Beans	15	7	7	7	14	8
Tomatoes	16	17	24	16	9	18

[a] A bean grown in the Philippines.

List of special foodstuffs issued on the average per individual per day, in grams.

Dried fish	7 to 8
Tinned salmon	10
Onions	125 to 130
Sugar	5 to 10
Macaroni	1.5 to 2
Chocolate	5
Condensed milk (cubic centimeters.)	5

Incidence of beriberi at Culion in 1909–10.

Month.	Total population.	Deaths.	Deaths from beriberi.	Total number.	In hospital.	
					Admitted sick.	Dismissed cured.
1909.						
January	1,170		6			
February	1,480		13			
March	1,430		15			
April	1'553		15			
May	1,575	47	14			
June	1,719	48	10			
July	1,770	87	8			
August	1,830	61	10			
September	1,776	65	21			
October	1,825	43	4			
November	1,761	80	31	33	1	1
December	1,807	188	68	70	1	1
1910.						
January	1,625	164	103	132	*23	6
February 1–8	1,600	26	9	9		

*These cases were examined on February 7 and 8, 1910, on an inspection trip to Culion.

The rice was changed in the beginning of January and at the same time the supply of the "mongo" [15] bean was increased.

While a sufficiency of data are not at hand to prove the cause of the outbreak, yet the great reduction in the number of cases after increasing the ratio of *mongo*, which is rich in phosphorus and protein, is striking.

At the suggestion of the writer, Dr. Victor G. Heiser, Director of Health for the Philippine Islands, in February, 1910, gave the order that only unpolished rice (pinaoa) should be supplied to the lepers in Culion. Beriberi disappeared among the lepers in April, 1910, and has not made its reappearance.

THE PHOSPHORUS CONTENT OF POLISHED AND UNPOLISHED RICE IN RELATION TO BERIBERI.

The next question to be considered is: Are these variations in the content of phosphorus as observed in the different kinds of rice sufficient to explain the difference in the action of polished and unpolished rice as foodstuffs capable or not of causing beriberi as well as polyneuritis gallinarum?

[15] *Mongo* is a small bean, *Phaseolus radiatus* Linn. (*P. mungo* Blanco), similar to *katjang idjo*, of Dutch India. The native physicians of this archipelago have proved it to be as valuable as the latter as a popular remedy for beriberi. This bean, according to my analysis, contains the following: Protein, 23.75 per cent; water, 9.56 per cent; P_2O_5, 0.77 per cent; fat, 4.5 per cent; crude fiber, 6.4 per cent.

No further samples of the November and December rice were obtainable, but that used from December 25 until January 12 and after this date has been analyzed and the following per cent of P_2O_5 obtained; rice from December 25 to January 12, 0.321; rice from January 12, 0.463.

From the standpoint of physiological chemistry, the process of milling the rice corresponds to that practised with other cereals and, therefore, the pericarp, removed by the process of milling, corresponds to what we term the bran of wheat or rye. In other words, white rice in composition corresponds somewhat to bread made from fine wheat flour, the red rice to that from whole wheat.

The importance of the bran from wheat and other cereals was formerly not very seriously considered, but more recently Jordan, Hart and Patten,[16] Hart and Andrews,[17] and others, have shown that an organic substance containing phosphorus and termed "phytin," the calcium-magnesium salt of phytic acid, is found therein. Phytic acid, or anhydrooxymethylenediphosphoric acid, was discovered by Posternak[18] as constituent of green plants, and several investigators have since then demonstrated its wide distribution in the vegetable kingdom. Hart and Andrews[17] found that practically all the phosphorus contained in vegetable foodstuffs is present as salts of phytic acid. This also holds true for rice. Suzuki, Yoshimura and Takaishi[19] proved that 85 per cent of the phosphorus in the bran of the rice is present as phytin. Phosphorus, combined in phytin in contradistinction to that in inorganic compounds, is readily soluble in water or dilute acids and can easily be removed from the bran by processes of washing or extraction. This solubility, as the investigations of Hart and his collaborators[20] have shown, is of great importance in relation to the behavior of phytic acid in metabolism. The phosphorus of phytic acid is readily absorbed, and is therefore a valuable compound in which to supply phosphorus in a form which can readily be assimilated. The effect of so-called organic phosphorus on metabolism in general has already been carefully investigated in the case of lecithin, and the superiority in many respects of organic phosphorus over certain inorganic phosphates seems to be proved.

The investigations, especially of Patten and Hart,[16] Mendel and Underhill,[21] Le Clerk and Cook,[22] proved that the same also holds true for the organic phosphorus compounds which are found in vegetables, namely, phytic acid and its salts. In addition to the readiness with which it is absorbed, phytic acid, according to the experiments of Patten and Hart,[16] is both a diuretic and a laxative, and the well-known laxative action of bran is partly produced by its content of this substance. On the other hand, these authors demonstrated in their work that constipation results if the bran is restricted or if a washed bran, poor in phosphorus, is ingested.

The hypothesis that lack of phosphorus may be an important factor in the etiology of beriberi gains in probability if we consider these facts as they have been found to exist in experiments on cattle, dogs and rabbits, and compare them with what we know concerning the action of different kinds of rice on human beings. For instance, our knowledge that the removal of phytin causes constipation fits well with the general

[16] *Amer. Journ. Physiol.* (1906), **16**, 268; *Amer. Chem. Journ.* (1904), **37**, 564.
[17] *Bull. New York Agric. Exp. Sta.* (1903), **No. 238.**
[18] *Compt. rend. Soc. biol.* (1906), **55**, 1190.
[19] *Bull. Coll. Agric. Tokyo* (1907), **7**, 495–572.
[20] *Amer. Journ. Physiol.* (1909), **24**, 86–103.
[21] *Amer. Journ. Physiol.* (1906), **17**, 75–88.
[22] *Journ. Biol. Chem.* (1907), **2**, 203.

observation that many beriberi patients suffer from extreme constipation.

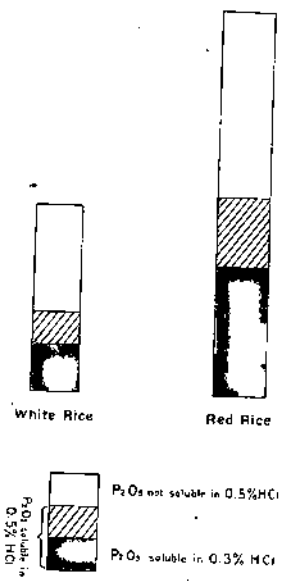
I have made comparative analyses showing the amount of "soluble" phosphorus in polished and unpolished rice and also in rice husk (*darac*) which is left as a result of the polishing process.

THE SOLUBILITY OF P_2O_5 CONTAINED IN WHITE AND RED RICE.

One hundred grams rice were extracted for five hours with 1,000 cubic centimeters 0.5 per cent hydrochloric acid. White rice gave 0.133 gram soluble P_2O_5 or 42.03 per cent of the total P_2O_5. Red rice gave 0.330 gram soluble P_2O_5 or 59.3 per cent of the total P_2O_5.

One hundred grams rice were extracted for two hours with 1,000 cubic centimeters 0.3 per cent hydrohloric acid. White rice gave 0.079 gram soluble P_2O_5 or 25.2 per cent of the total P_2O_5. Red rice gave 0.221 gram soluble P_2O_5 or 39.7 per cent of the total P_2O_5.

The following is a graphic representation of these results and shows them very plainly.



The solubility of the P_2O_5 in rice husk obtained by polishing (darac).

Fifty grams husk, containing 8.27 per cent water, were treated for five hours with 300 cubic centimeters of the solvents given below:

Per cent dissolved by—	P_2O_5.	Nitrogen.
Cold water	0.34	0.269
Hot water	1.70	0.314
0.25 per cent HCl cold	1.66	0.326
0.5 per cent HCl cold	2.04	0.407
1.25 per cent HCl cold	2.25	(0.312)?
Total	4.28	2.00

These analyses strikingly demonstrate not only that the total amount of phosprorus in unpolished rice is considerably higher than in polished, but also that a greater percentage of the total phosphorus in the red variety than in the white is in a readily soluble condition. Therefore, if the same quantity of red as of white rice is eaten, twice the total quantity of phosphorus and nearly three times as much of the soluble phosphorus compounds will be ingested. (See diagram.)

I next endeavored to ascertain if lack of phosphorus, and especially if that in the soluble form, is the cause of the deleterious action of white rice. If this is correct, then the addition of organic phosphorus (phytin) to white rice should render the latter harmless.

I began with experiments on fowls, following the method of Eykman, and convinced myself that chickens fed on white rice contract polyneuritis whereas control birds fed on unhusked and even white rice, if the latter is carefully mixed with rice bran (*darac*), remain healthy for two and a half months.

It is not my intention to prove to what extent the polyneuritis of Eykman and the disease which we term beriberi in man are related, or even to decide whether or not they are one and the same; but I believe these experiments give us a basis of investigation in order to ascertain the importance of the lack of certain food constituents (such as phosphorus) in producing pathologic changes in the nerves and other tissues.

In pursuance of this field of observation, I fed two groups of chickens; the one on white rice and phytic acid prepared from rice husk; the other on white rice alone. The phytic acid was isolated in the following way from the bran of white rice which is separated by the process of polishing between the rotating stones of the mill:

> Four kilos of this material (native name *darac*) were placed in a small barrel and extracted with 6 liters of 0.3 per cent hydrochloric acid for from four to six hours, the mixture being stirred from time to time. The extract was filtered, sodium acetate added to the filtrate, and the phytic acid precipitated as its copper salt by means of copper acetate. The phytate of copper was separated, washed with small amounts of water, suspended in this liquid and the copper precipitated by hydrogen sulphide, the precipitate separated by filtration and the filtrate evaporated on a steam bath. A brownish, highly acid sirup was obtained in this way. Its reactions, as well as the manner of its preparation, identified it as phytic acid, according to Patten and Hart.[10]

The free acid was used in the first series of experiments. The rice to be fed was either cooked or only mixed with about 10 cubic centimeters of a solution of phytic acid which corresponded to 0.2 gram of P_2O_5. At first the birds ate this rice very well, but great difficulty in inducing them to take enough of the prepared rice arose subsequently. In a second series of experiments, I therefore endeavored to improve the attractiveness of the preparation of phytin, at first by adding sugar; but

as this did not increase its palatability I neutralized the acid by means of sodium or potassium hydroxide. But the alkali salts, while they did not offer as great an obstacle, were also not attractive to the birds. Finally, I prepared calcium phytate by neutralizing a concentrated solution of phytic acid in water by means of a concentrated solution of calcium acetate. The calcium salt of phytic acid is not very soluble in water; it is precipitated and filtered. The preparation has but a slight taste and the odor of acetic acid. In the last two months of the second set of experiments, this preparation was fed directly, each fowl receiving daily in the morning one gram of the calcium phytate.

All the birds which I used were young, still growing, and from about 600 to 1,000 grams in weight. I chose young chickens because I believed that they would contract the disease at an earlier period than old ones. The rice was at first given almost in the dry state, and later, as a change, cooked rice was also prepared, but the birds seemed to take even less of the latter than of the former. A sufficient quantity of the grain was given each day, and care was taken that some rice was always left over. It seems worthy of note that the animals at first instinctively attempted to pick out the grains of rice which were not so highly polished, and if a mixture of white and red rice was given, they would select all the unhusked grains, leaving the others. My general observation is that chickens in the beginning will take white rice in large amounts, but after some time they tire of it and then they do not eat it readily. The course of the experiments is given in the following tables:

EXPERIMENT I.—*Fowls fed on white rice and white rice with phytic acid.*

Five (six) chickens kept in large wooden cages.

WHITE RICE.

Mark of fowl.	Month, day, and weight in grams.					Beginning of the disease.	Death.
	Sept. 13.	Sept. 20.	Sept. 27.	Oct. 3.	Oct. 8.		
a	575	464	431	394	351	Oct. 9	Oct. 14
d	469	421	323	319	305	Oct. 8	Oct. 10
e	314	300	233	224	236	Oct. 12	Oct. 15

WHITE RICE AND PHYTIC ACID.

Mark of fowl.	Month, day, and weight in grams.							Result.
	Sept. 13.	Sept. 20.	Sept. 27.	Oct. 3.	Oct. 8.	Oct. 15.	Oct. 21.	
b	500	Escaped Sept. 14.						
c	417	456	439	479	522	505		Transferred to experiment II.
f	340	331	317	288	298	272	253	Oct. 25, died showing symptoms of paralysis beginning Oct. 26.

EXPERIMENT II.[a]—*Fowls fed on white rice, white rice and darac, and white rice and phytic acid.*

Eight (nine) chickens kept in a large yard inclosed by a wire fence.

WHITE RICE.

Mark of fowl.	Beginning of disease.	Death.
g	Nov. 23	Nov. 26
h	Nov. 11	Nov. 13
k	Nov. 9	Nov. 20

WHITE RICE AND DARAC.

Mark of fowl.	Beginning of disease.
l	Healthy until Dec. 31
m	Do.

WHITE RICE AND PHYTIN.[b]

Mark of fowl.	Beginning of disease.	Death.
c (from foregoing experiment)	Healthy until Dec. 31	
e	Dec. 23	Dec. 26
i	Nov. 1	Nov. 18
j	Healthy until Dec. 31	

SUMMARY.—*Duration of life (days).*

Chickens receiving white rice only.		Chickens receiving white rice and phytic acid.	
Mark of fowl.	Days.	Mark of fowl.	Days.
a	33	c	> 120
d	30	f	44
e	34	c	76
g	46	i	38
h	33	j	> 80
k	40		

[a] This experiment was commenced October 12, 1909, and ended December 31, 1909.

[b] October 13 to October 25, 10 cubic centimeters of phytic acid solution were given daily (0.2 gram P_2O_5); October 26 to October 31, the same solution of phytic acid neutralized by caustic potash; November 1 to December 30, 1 gram calcium phytate; November 7 to November 10, owing to illness of the writer, no phytin was given, but white rice only.

All chickens fed for some four to five weeks on white rice developed the typical symptoms of polyneuritis[28] as they have been described by Eykman and other investigators. These are very characteristic and scarcely need pathologic-anatomic confirmation. The weakness of the legs and the characteristic gait of the birds are sufficient to confirm the diagnosis.

It is evident from these experiments that chickens which are given white rice alone do not live much over thirty-five days, whereas the birds fed on white rice plus phytic acid live for a considerably longer time. One fowl from the first group which received phytic acid, survived for three months, being under observation continually, whereas a second bird fed on white rice together with phytic acid contracted the disease and died, but it did so at a later time than the control chickens fed on white rice only.

In the second set of experiments, I succeeded by the addition of the calcium salt of phytic acid in keeping two chickens alive for nearly three months on white rice only. However, one chicken already became ill after 3 weeks and died on the 38th day. I can not state the exact reason for this result, but I believe it to be true that the bird did not take enough phytin in the beginning of the experiment in which fowls were fed on a mixture of rice and phytic acid and at which period they were more or less able to select those grains of rice which were comparatively not as thoroughly mixed with the preparation.

We may draw the conclusion that without doubt phytic acid renders the effect of a diet of white rice less damaging. I did not have as great success by the use of phytic acid as in feeding red rice, or by adding *darae* (rice husk), neither was the protective effect as great as that described by Eykman when he gave red rice, or added beans or meat to the diet.

Schaumann,[13] in his latest communication, which came into my hands for the first time after I had finished the experiments described above, shows that he was able to protect pigeons from the damaging effect of white rice by the addition of yeast or wheat bran. These experiments increase the number of substances which we already know and which act as a protection against polyneuritis produced by feeding white rice. However, all of these other experiments do not prove *which* constituent of

[28] The histologic examination of the *nervi ischiadici* and *tibiales* of these animals and of those described farther on in this paper was kindly undertaken by Dr. Vernon L. Andrews, of the pathological laboratory of the Philippine Medical School. The sections showed plainly a number of degenerated fibers in the nerves.

the substances to be added counteract the damaging effect of the white rice. I believe my experiments give sufficient proof *at present* that at least *one of the main factors* in these protective substances must be an organic compound of phosphorus, because of the fact that the addition of such organic phosphorus, as it is present in the rice husk, has a protective action against the damaging effect of the white rice itself. I agree with the interpretation which Schaumann has given to his own experiments and to those of Eykman. He may be correct in considering that the nucleo-protein of yeast is the constituent preventing polyneuritis, but in the case of rice it is my belief that the phosphorus compound which has been designated as "phytin" is the active substance.

Although my experiments seem to be sufficiently positive to warrant a continuation along the same line, I have abandoned them at present in order to discover if it is not possible to work with animals other than chickens. These birds are not very appropriate for exact experimentation, and while we can endeavor to produce in them such diseases as polyneuritis, it is very tiresome to carry on close feeding experiments with fowls. I have therefore attempted to feed other animals on white rice or on a food having a similar composition.

Monkeys were fed on white bread made only of wheat flour and water and salt. Such a bread, when fresh, contains about 10 per cent of water, 8 of protein, 52 of carbohydrates, 0.116 of P_2O_5; or, if we reduce the percentages to the same water content as rice, about 10 per cent of protein, 75 of carbohydrate and 0.155 of P_2O_5. Therefore, this bread in its P_2O_5 content corresponds to a very highly polished rice; the protein content is slightly higher, that of carbohydrates somewhat lower, but these differences are really too small to be of any importance.

The animals in the beginning took this bread readily. At first I gave 75 and then 100 grams per monkey of 2,000 grams; this amount being almost entirely eaten up each day, up to the time, when the monkeys became very weak. This amount of bread corresponds to 100 calories per kilo of body weight. It must be remembered that the demand for energy in these animals is very great when we consider their small size, great surface and their excessive muscular activity. The animals (*Macacus philippinensis* Geoff.) were kept in a large, airy yard, covered on top and on one side with wire gauze, thus giving them plenty of air and more space for free movement and gymnastic exercise than is found in the usual cages. Furthermore, the animals were placed at least two in a room so that they would not feel their imprisonment quite so keenly. In working with monkeys this point must not be forgotten.

EXPERIMENT IV.—*Weight of monkeys fed on bread and water.*

Date.	I.	II.	III.	IV.	V.
January 10	1,853	2,083	2,123		
January 17	1,324	[a] 1,814	2,131		
January 25	1,315		2,087	1,745	1,440
February 1	1,287		2,102	1,722	1,447
February 8	1,077		2,037	1,590	1,430
February 15	1,079		2,005	1,480	1,400
February 23	1,000		1,975	1,545	1,375
February 28	920		1,720	1,530	[b] 1,130
March 1	[c] 842				
March 2			[d] 1,745	(e)	

[a] Died January 18, strong diarrhœa, unknown origin.
[b] Died February 27, weakness, etc., as described below.
[c] Died March 1, weakness, etc., as described below.
[d] Died March 2, weakness, etc., as described below.
[e] Died March 26, weakness, etc., as described below.

Three animals were selected at first, but one (Number II) died in the beginning of the experiment of a severe diarrhœa of unknown origin. Two others were taken about two weeks later. The experiment demonstrated that bread and water alone are not capable of sustaining monkeys in full health for a longer period than from four to six weeks. In spite of the fact that the animals ate the equivalent of 80 to 100 calories and 0.3 gram protein per kilo of body weight, they died in about six weeks, with signs of general weakness and emaciation. The muscular weakness in some of the monkeys was very prominent. The animals, which in the beginning would run away if approached and if caught would resist their captor with considerable muscular force, were now handled without trouble and were scarcely able to hold themselves with their hands gripping the arm or fingers. Death came at last, the respiration becoming slower and slower, and weaker and weaker. At autopsy no pathologic changes other than those incident upon emaciation and anæmia were noticed. The histologic examination of the nerves has not up to the present been undertaken. No experiments on the action of phytin on monkeys nourished in the manner outlined above were made.

Guinea pigs will not eat white rice, either cooked or uncooked. When given it they die of starvation in from two to three weeks' time.

I attempted, but with considerable difficulty, to feed adult dogs on white rice and lard. A series of experiments on young dogs will be described later, in connection with other work.

Schaumann,[13] in his latest paper, reports that he was able to produce lesions of polyneuritis in dogs, cats, and rats by feeding them horse meat which had been heated in a 20 per cent solution of sodium carbonate for three hours. However, the full details have not as yet been published.

It seems not out of place to mention here some other work which may have an important bearing on the question under consideration.

Quite recently Stepps[24] has shown that mice when fed on a bread made of flour and milk, which had been extracted with ether or acetone and so deprived of the lipoids, died in a few weeks, while they survived if given the unextracted bread or the extracted bread plus the extract. The author emphasizes the absence of fat after this process, but I think that the extraction probably removes the organic compounds of phosphorus (lipoids) and the loss of these bodies makes the bread unfit for food. The action of food poor in phosphorus on young animals has been studied in the last year by Hart, McCollum and Fuller[25] and by Heubner and Lipschütz.[26] In both publications the authors state that pathologic lesions of the skeletal system are the main results of the taking of food lacking in phosphorus, but, in addition, Heubner and Lipschütz report that they have observed nervous symptoms in one of their dogs, and they mention the possible connection of this question with beriberi.

CONCLUSIONS.

If we summarize our experiments and the observations recorded in the literature, we can state as follows:

1. Certain foodstuffs, especially rice, which are relatively poor in phosphorus (phytin) if they are the main or exclusive article of diet for any great length of time, have been shown by various authors to cause beriberi.

2. The process of polishing removes the outer layers from the rice; these are rich in phosphorus, especially soluble organic compounds of that element (phytin). They are food constituents probably of high physiologic importance.

3. A diet similar to that which is regarded as the probable cause of beriberi if exclusively given for any length of time to animals, is not sufficient to keep them in normal health.

4. Polyneuritis has been observed in chickens receiving a food similar to that which is regarded as causing beriberi, namely, white rice; the addition of organic phosphorus in the form of phytin or its salts considerably, but not entirely, reduces the deleterious effect of a diet (white rice) which can produce polyneuritis in chickens.

[24] *Biochem. Zischr.* (1909), **22**, 452–460.
[25] *Amer. Journ. Physiol.* (1908), **23**, 246–277.
[26] *Verhandl. 26 Vers. Gesellsch. f. Kinderheilk.* (1909), 149–161.

PHOSPHORUS STARVATION WITH SPECIAL REFERENCE TO BERIBERI: II.

By Hans Aron and Felix Hocson.

In order to reach a definite conclusion concerning the questions under consideration, it is necessary further to prove that phosphorus starvation actually takes place when a man lives on a diet consisting only of polished rice, or on one of similar composition, and that such starvation does not take place if unpolished rice is used to the exclusion of the other variety.

The minimum amount of phosphoric anhydride in the daily food allowance, according to Tigerstedt,[27] Ehrströhm,[28] and Renvall,[29] should be about 3.4 grams of P_2O_5 daily for an average man, namely, 0.06 gram per kilo of body weight.

Now, if we consider the quantity of phosphorus taken in food, the main constituent of which is white rice, this amount will appear to be exceedingly low as compared with that present in a normal European diet.[30]

However, if we take the values given by Ehrströhm and others as normal, then every Filipino living in his accustomed manner should be nearly in a state of phosphorus starvation; for even a diet of fish and rice scarcely furnishes two grams of P_2O_5 daily, which is less than 0.04 gram per kilo of body weight.

Other authors [34] believe the figures given by Tigerstedt and Ehrströhm to be too high and contend that about 0.035 gram P_2O_5 per kilo body weight is sufficient. We quote the following data taken from a publication

[27] *Handbuch d. Phys. Braunschweig* (1908), 1.
[28] *Skand. Arch. f. Physiol.* (1903), 14, 91.
[29] *Ibid.* (1904), 16, 94–138.
[30] *Deutsches Arch. f. klin. Med.* (1899), 63, 386 422.
[31] Meyer, L. F. *Ztschr. f. phys. Chem.* (1904), 43, 1–10; Magnus-Levy, V. Noorden Handbuch der Pathologie des Stoffwechsels. Berlin, 2d ed. (1906), 1, 457.

by Oeri;[32] these show that a quantity less than 1.5 grams of P_2O_5 daily would doubtless be insufficient to fulfill the demands of the body.

Author.	P_2O_5 intake per day.	P_2O_5 in urine.	P_2O_5 in fæces.	P_2O_5 balance.
Sivén	1.023	0.999	0.677	−0.653
	1.464	0.983	0.828	−0.347
	3.545	2.039	1.050	+0.460
Hämäläinen and Hehue	1.185	0.979	0.559	−0.354
	1.271	0.919	0.463	−0.112
	1.277	0.902	0.585	−0.209
	1.227	0.814	0.534	0.122

The question as to the minimum amount of phosphorus which is necessary is probably dependent upon conditions very similar to those attending the problem of the minimum amount of protein. The variety and character of the phosphorus compounds taken and certain other conditions inherent in the individuals are probably the determining factors. However, it seemed to be necessary to ascertain if the amount of phosphorus taken by the Filipino in his typical diet is sufficient in quantity, and also how far this amount of phosphorus can be reduced without producing a state of phosphorus starvation; and, further, it seemed necessary to investigate the bearing on metabolism of a diet low in phosphorus, namely, of polished rice, or a similar one richer in this constituent, namely, unpolished rice.

It seemed to us, in undertaking such experiments, very important not only to study the phosphorus, but also the nitrogen metabolism, for the following reasons: A diet known to cause beriberi, consisting almost entirely of rice and deficient in meat, fish, and other substances, is also relatively poor in protein, and it furnishes the latter only in the form derived from plants. Proteins from plants are not as fully digested as those from animals, and, furthermore, as certain quite recent investigations [33] indicate, vegetable protein has not the same value for the body as an equal amount from animals. Finally, metabolism experiments, especially those of Le Clerk and Cook,[34] demonstrate that the addition of organic phosphorus increases the retention of nitrogen, an observation which is in accordance with statements frequently made [35] regarding the close connection between nitrogen and phosphorus metabolism in general.

[32] *Ztschr. f. klin. Med.* (1909), **67**, 288–306.

[33] Thomas, Carl. *Arch. f. Anat. u. Physiol.* (1909), 219–302; Michaud, *Ztschr. f. physiol. Chem.* (1909), **59**, 405–491.

[34] *Journ. Biol. Chem.* (1907), **2**, 203.

[35] Tunnicliffe. *Arch. int. d. Pharm. u. Ther.*, **12**, 207.

METABOLISM ON A DIET POOR IN PHOSPHORUS WITH AND WITHOUT THE ADDITION OF THE ORGANIC PHOSPHORUS COMPOUNDS PRESENT IN THE RICE BRAN SEPARATED BY POLISHING.

NORMAL MAN.

Following out the above considerations we first undertook a series of metabolism experiments on normal men, in order to determine the intake and outgo of phosphorus and nitrogen of Filipinos kept on the usual diet, which is low in phosphorus, with and without addition of organic phosphorus in the form of rice bran (rice polish) and of phytin itself. We also, finally, because of the above-mentioned reasons, studied the effects of such a diet, with and without the addition of protein.

For these experiments and for those described later on, we used prisoners in Bilibid Prison, who voluntarily submitted themselves to the changes in diet. We are very much indebted to Doctor Christensen for his kindness in permitting us to use the facilities of Bilibid Hospital and Dr. Pineda for his kind help in the observations. The conditions there existing for carrying on metabolism experiments are very good, and the persons under observation could be kept without any trouble under lock and key, in a quarantine room with cement floor and walls, furnished with only a bed and small table. A chemical balance and the apparatus for collecting urine and fæces were added.

Two normal men were used as subjects for experiment. No. 1 had undergone an operation for hernia three weeks before he subjected himself to the feeding experiments, but had fully recovered. A comparative study was made with a diet not as rich in phosphorus as is usual and with the same diet to which rice bran from polishing had been added. At first we endeavored to feed the man almost entirely on rice, allowing, in the first and second periods, sugar, salt, and coffee as the only additional foodstuffs. However, he rejected his food several times and asked for some fish and other things. Therefore it was necessary to interrupt the experiment at this point and to give a diet of rice, bread, sugar, coffee, and fish. Bread in reality is not a typical constituent of the diet of Filipinos, but we used it because it does not require cooking and therefore simplifies the preparation of the experimental diet. It must also be noted that the bread was white, made of wheat flour and water after the usual manner, and low in phosphorus and protein, but rich in carbohydrates. In this respect it is very similar to highly polished rice.

The rice was of the white variety, but only slightly polished; the fish was one of the larger native species, half-dried and smoked; the bones were carefully removed and the remainder cut into small slices. These pieces were thoroughly mixed, to secure average samples, and 8 portions of 40 grams each were weighed out and kept on ice. A sample was at the same time taken for

analysis. Sugar 50 respectively 100 grams, rice 300 (250), bread 300 (250), and coffee 300 cubic centimeters were freshly weighed or measured each day. The rice was all from the same stock and an average sample of it was taken for analysis. Samples of the bread weighing 50 to 100 grams were taken every second day and preserved in formalin. All the samples were finally dried, weighed again, mixed and finely powdered. Coffee secured on several days was mixed and also analyzed. The sugar was fairly pure, and no analyses were necessary. The *darac* or rice husk from the polishing process, which was used in the second period, was heated to 100° for twenty-four hours to destroy organisms which are always present. Four parts, of 75 grams each, were weighed out and a sample was taken at the same time for analysis. In calculating the amount of the *darac* which it was necessary to add, we regarded the bread and rice given in total as "white rice;" and corrected for both, by addition of the bran to "red rice;" 50 grams of bread and 50 grams of rice being replaced by 75 grams *darac*, and, in order to equalize the calories which had been removed from the diet, we increased the sugar ration from 50 to 100 grams. The man received three meals a day: Breakfast at 7 a. m., bread, coffee and sugar; luncheon, 11.30 a. m., rice and fish, with salt *ad libitum;* supper, 5 p. m., bread slightly toasted, coffee, sugar.

The experiment was carried on without difficulty. The unsuccessful days of the first and second period were sufficient to train the man and his attendants carefully to collect the urine and fæces, and to consume completely the food given; so, for example, he removed all remaining particles by licking the plates.

The urine was placed in a pear-shaped receptacle and then poured into a stoppered bottle containing 20 cubic centimeters diluted formalin. In later experiments the urine was preserved with thymol, formalin giving a certain amount of precipitate which made it difficult thoroughly to mix the urine for analysis. Urine was collected every twenty-four hours, from 8 a. m. to 8 a. m., the total quantity was measured and a known fraction was preserved. In subsequent experiments the quantity was measured, and, by the addition of water, it was diluted to an even amount, such as 1,500 or 2,000 cubic centimeters, so as to simplify the preparation of the mixed urine. The fæces were deposited in a large museum jar, closed by a glass cover and containing 100 to 150 cubic centimeters of formalin, a sufficient quantity to deodorize and preserve the excreta. Not infrequently more formalin was added on the second or third day, so as better to penetrate the scybala with the antiseptic. This method, while it does not permit the determination of the total quantity of *fresh* fæces, proved to be very satisfactory under the present conditions, where the laboratory and the experimental subject were so great a distance apart.

The fæces for each period were marked off by carmine, a method which, except in a few instances, gave very satisfactory limitations. An aliquot portion of the urines (one-tenth of the daily amount) from each period was mixed, provided the analysis of an individual urine was not demanded for special reasons.

The total amount of fæces plus water and formalin was determined for each period by weight, and as large a fraction as possible (one-half to two-thirds of the total amount) was dried in a weighed porcelain dish, under addition of sulphuric acid, on a steam bath. The drying process was finished in a steam-heated vacuum-drying apparatus, then the dish with the fæces was exposed to the air for one day, the quantity of air-dried material determined and then

finely ground in a mortar, the powder being kept in a closed bottle. Determinations of total nitrogen (Kjeldahl) and phosphorus (Neumann's method) were performed on all samples of foodstuffs, urines and fæces. The somewhat complicated apparatus described by Neumann was not used for the destruction of the organic material, but a simpler device of my own design was employed.[30] The substance to be analyzed was placed in a 750 cubic centimeter Jena round flask with a long neck, 10 to 15 cubic centimeters of sulphuric and nitric acids were added at the beginning, whereas when the digestion approached completion, nitric acid alone was added.

The detailed records of the experiments are given at the end of this paper.

The subject during the first experimental period (III) of four days received 10.99 grams nitrogen and 1.67 grams phosphorus daily; he excreted 10.61 grams nitrogen and 1.66 grams phosphorus per day, so that nitrogen and phosphorus equilibrium practically existed, thus demonstrating that the food as given, representing about 1,900 calories and containing 70 grams of protein per day was sufficient to maintain a man weighing 52.5 kilos and performing practically no work. The intake of phosphorus, 1.67 grams or 0.032 gram of P_2O_5 per kilo of body weight, was absolutely sufficient. This amount is doubtless lower than the figure obtained as a result of experiments on Europeans, which amount has been regarded almost as the minimum amount of phosphorus permissible; but it corresponds approximately to the quantity of phosphorus demanded by dogs per kilo of body weight which, according to the experiments of Meyer,[31] is 0.035 gram only.

During the second period (IV) the man received very nearly the same diet, with the difference that bran from polished rice was added. He took 11.49 grams nitrogen per day, of which 9.61 grams were in the same form, namely, as rice, bread, and fish, as in the foregoing period, whereas 1.88 grams were present in the *darac*. The quantity of nitrogen taken is slightly higher, but it was necessary to assume that the nitrogen compounds in the *darac* are less digestible than those in the other foodstuffs. The quantity of phosphorus (5.46 grams) was more than three times as great as during the first period, but only 1.45 gram of the phosphorus was present in the same foodstuffs as were employed in the first experiment and 4.01 grams were in the *darac* (rice bran).

The outgo of nitrogen was 10.92 grams; that of phosphorus, 4.84 grams; the nitrogen balance was only slightly higher in this experiment than in the first one, but a retention of more than 0.5 gram phosphorus per day took place. The observation is very common that during the taking of a diet rich in phosphorus small amounts of the latter are retained.

The relative distribution of nitrogen and phosphorus in the urine and fæces was as follows: In the first portion of the experiment 24.06

[30] Handbuch der biochemischen Arbeitsmethoden. Berlin and Wien. (1909), 1, 388.

per cent of the nitrogen and 31.5 per cent of the phosphorus given reappeared in the fæces. A 75 per cent absorption of nitrogen corresponds to the average obtained with a diet more or less of a vegetable character.

McCay,[37] in his metabolism experiments on Bengalis, observed that 23.86 to 25.68 per cent of the nitrogen reappeared in the fæces. His subjects were given a very similar diet, principally vegetable in nature.

In the second part, 32.55 per cent of the nitrogen reappeared in the fæces. If we assume the absorption of nitrogen (and phosphorus) present in the mixed food exclusive of the *darac* to be the same in this part of the experiment as in the preceding, then it follows that 76.7 per cent of the nitrogen in the *darac* was excreted and only 23.3 per cent absorbed. According to this calculation also, the conclusion can be drawn that only 25 per cent of the phosphorus from the *darac* appeared in the urine and 75 per cent in the fæces.

I intentionally do not use the word "absorbed," because we know that the quantity of phosphorus excreted in the fæces is by no means a measure of the amount not absorbed, for a large proportion of the phosphorus is absorbed and *reëxcreted* into the fæces, its distribution between fæces and urine depending much more on other factors than on the absorbability of the phosphorus.

Oeri[38] in a recent set of experiments has shown that the proportion of calcium in the food is a factor determining the distribution of phosphorus in the excreta, urine or fæces. Patten and Hart also, in their experiments on cows fed with wheat bran, have found that the phosphorus of the wheat bran is excreted almost entirely in the fæces, but at the same time they demonstrated that this constituent in the fæces must previously have been absorbed; it is present in the bran as *organic* phosphorus, but is transformed into and excreted as inorganic phosphorus. As this process can not be performed by the digestive enzymes alone, it is more than probable that phosphorus in the form of organic compounds such as phytin is absorbed and reëxcreted into the fæces.

The next step in our investigation was to institute a comparative study of a diet lower than the first both in phosphorus and in nitrogen, and further to study the effect of an addition of organic phosphorus in the form of phytin to it. This portion of the experiment was also conducted as a control to metabolism experiments on a beriberi patient, to be described later. For this reason, as well as to learn the effect of a higher intake of protein, an addition of egg albumen was made in one of the experimental periods (VI). Another prisoner, No. 17794, was selected for the purpose. This man previously had suffered from a slight panaritium on his foot, but had fully recovered at the time the experiment began. He was strong and muscular, and did not spend approximately the entire time lying in bed, as did the first subject, but

[37] *Sci. Mem. Off. Med. San. Dept. India*, Calcutta (1908), 34, 1–67.

cleaned his room and did other things. The experiments on this man were divided into three periods, each covering four days. In the first, a diet low in phosphorus was given, in the next, egg albumen, and in the third, phytin was added. This man, weighing 64 kilos, received 400 grams of bread, 300 grams of rice, 75 grams of bacon and 100 grams of sugar daily, thus giving him 41 calories per kilo of body weight. More detailed information concerning the foodstuffs, etc., will be given under the head of the next experiment, which was in part conducted simultaneously with this one. Both men were naturally kept in two different isolation rooms.

A study of periods V to VII shows that in the first period where the man received bread, rice, bacon and sugar only, and took in daily 9.57 grams of nitrogen and 1.50 grams P_2O_5, the intake of nitrogen as well as that of P_2O_5 *did not cover* the needs of the body. The quantities 0.023 gram P_2O_5 and 0.15 gram nitrogen per kilo of body weight are therefore insufficient. The increase of nitrogen in the diet from 9.58 to 11.06 grams by the addition of egg albumen (period VI) reduced the loss of nitrogen considerably, namely, from 4.67 to 1.98 grams per day.

The loss of P_2O_5 at the same time is slightly decreased, as compared with the foregoing period. While the first man, weighing 52 kilos, with 11 grams nitrogen per day was exactly in nitrogen equilibrium, the other, of 64 kilos weight, with the same intake, lost nearly 2 grams of nitrogen. The absolute requirement of both men would therefore amount to approximately 0.2 gram of nitrogen per kilo of body weight.

Finally, during the last period (VII), with the same intake of nitrogen as in period V, but a large increase in that of P_2O_5, brought about by the addition of 6 grams of phytin daily, 1.5 grams P_2O_5 were retained daily from the amount given; this retention probably being caused by the fact that more than 5 grams of P_2O_5 had been lost from the body during the two foregoing periods (V and VI).

Special attention is called to the nitrogen metabolism. Whereas the intake in period VII is about the same as in period V, the loss of nitrogen from the body is not half so great. This influence of the organic phosphorus upon the nitrogen metabolism is in accordance with the observations of Mendel and Underhill,[21] Le Clerk and Cook,[24] Tunicliff [25] and others, mentioned above. It shows that nitrogen metabolism must not be forgotten while studying that of phosphorus under different conditions of nutrition.

The distribution of nitrogen and P_2O_5 in the urine and fæces during these experiments was as follows: The content of the fæces in nitrogen is nearly the same during the three periods, being about 3.0 to 3.4 grams. This amount would accord with nearly 35 per cent of the intake corresponding to the periods without the addition of protein (V and VII). The values would agree fairly well with those found by other authors

for a purely vegetable diet.[38] During period V the output of nitrogen in the urine alone is greater than the amount taken in. In all this series of experiments the quantity of phosphorus which appears in the fæces is somewhat higher than in the foregoing, but this is scarcely regarded as of importance. The fact that in period VII all the phosphorus from the phytin so far as it is again excreted, reappears in the fæces seems to me deserving of more attention, the quantity of phosphorus in the urine not being increased at all. The explanation given above for this excretion of phosphorus through the fæces probably also holds good. (See p. 103.)

Our conclusions, deduced from the above experiments in their relation to beriberi, are as follows:

(1) A diet consisting of bread and rice (both poor in phosphorus), some fat (bacon) and sugar, furnishing 40 calories, 0.15 gram N and 0.025 gram P_2O_5 per kilo body weight does *not* cover the demands of the body for N and P_2O_5 and therefore leads to N and P_2O_5 loss from the body. Addition of protein reduces the N loss of the body and the loss of P_2O_5 slightly.

(2) The addition of phosphorus in the form of phytin prevents a loss of that constituent from the body, and if sufficient of this element is added a storage of phosphorus after a period of phosphorus starvation takes place. The loss of nitrogen from the body is reduced by the addition of phytin, as compared with a corresponding period in which phytin is not given.

(3) A diet consisting of fish, bread, rice, sugar, etc., furnishing 37 calories, 0.2 gram of nitrogen and 0.032 gram of P_2O_5 per kilo of body weight, is sufficient to keep a man in nitrogen and P_2O_5 equilibrium.

(4) The addition of rice bran has a tendency to produce a slight storage of P_2O_5; the rice polish in this respect corresponding to phytin. The phosphorus, both of rice and of phytin, is excreted almost entirely in the fæces.

METABOLISM EXPERIMENTS ON A BERIBERI PATIENT.

We next attempted to study the metabolism of a beriberi patient under the same conditions. The work so far done in this connection is very limited.

Schaumann,[9] in conjunction with his researches, states that the urine in cases of beriberi has a very low content of phosphorus, and Durham[39] found the metabolism in beriberi to be depressed, the urine having a low content of urea, phosphates, etc. I do not believe that these facts are of very great importance, because we know nothing concerning the food taken at the time and its content of phosphorus.

Scheube[40] also has made a number of analyses of urine in cases of beriberi and comes to the conclusion that in this disease metabolism is lowered.

[38] Yukawa, Genyo, *Arch. f. Verd.-Krank.* (1909), 15, 477–524, 609–646.
[39] *Brit. Med. Journ.* (1904), 2, 27.
[40] *Deutsches Arch. f. klin. Med.* (1882), 31, 141, 307.

Teruuchi and Saiki,[41] in opposition to Durham, as a result of an experiment on metabolism, state that the destruction of protein is *increased* during beriberi. Finally, Miura,[42] in a recent article on this disease which came to my notice only after this paper had been read before the association, quotes a number of experiments on metabolism which he has made on 4 people suffering from a more or less acute attack of beriberi. It is characteristic that all of his patients were undernourished during this time. The caloric intake per day often was not higher than 250 to 300 calories, on an average not 1,000. The loss in body weight during the short experimental periods was correspondingly high, namely, 55 to 49, 55 to 46, 46 to 42 kilos, etc. He concludes: "In acute, severe beriberi the nitrogen and P_2O_5 excretion is increased in the urine independently of the quantity of urine passed, in subacute beriberi the nitrogen loss is much lower." His results are very much obscured by the fact that his patients were at the same time highly undernourished.

It was our good fortune to encounter one case of typical beriberi in Bilibid Prison which we could use for study of metabolism. The disease is not at all common among the prisoners in this institution; if the diet given to these people and upon which one of us reported [43] during the past year is considered, it is scarcely to be understood how beriberi could be present at all among its inmates. Indeed, every case occurring there would argue against the theory that beriberi is caused by a diet low in phosphorus, unless a reasonable explanation could be produced showing that the man suffering from beriberi had not received the ample and healthful diet of Bilibid Prison. The case which we encountered, prisoner No. 7272, can easily be proved not to have had a full diet just before he contracted the disease.

The records show that this man has been in prison from April 30, 1908, up to the present time, serving a life sentence. Up to September 25, 1909, his conduct was sufficiently good so that he had suffered only light punishment, such as "carrying stone." During this period his full diet was never restricted, but on September 25, 1909, he received his first severe punishment, he being condemned to ten days on *bread and water*.

His second punishment began on October 22, 1909, and from this date up to December 22, 1909, he was placed on bread and water for a total of forty-one days, and on one day he received two meals, consisting only of bread and water.

A summary of these dates and conditions is as follows: From September 25 to December 22 (a total of eighty-eight days), he was on a diet of bread and water during fifty-one and two-thirds days; or, from October 22 to December 22 (a total of sixty-one days), he was on *bread and water* for forty-one and two-thirds days.

On December 22 this prisoner was taken to the hospital with typical symptoms of beriberi. From this time on he was kept in the hospital on a mixed diet and the Filipino assistant, Dr. Pineda, who treated him attempted to induce him to take *mongo;* however, the prisoner had no

[41] *Mitt. med. Gesellsch.* Tokyo (1905), **19**, No. 6.
[42] Beriberi oder Kakke. *Ergeb. d. inn. Med. u. Kinderheilk.* (1910), 4, 280–318.
[43] Aron, *This Journal, Sec. B* (1909), **4**, 195.

taste for this food and therefore, when we first saw him, in the beginning of January, he was practically on a diet of rice, together with a certain quantity of fish and meat, milk and bread. The results of the examination are as follows:

A Filipino of small stature, 42 kilos in body weight, lying in the bed, unable to raise himself or to stand without assistance. If placed on his feet, he collapses as soon as support is released. Muscles of the leg and arms very weak, knee reflexes absent; dynamometric test of the left hand 30 to 35, right hand 55 to 60. The æsthesiometric test shows that the ability to feel, the sense of touch and reaction to pricking, as well as his ability to distinguish two different spots simultaneously touched, is reduced to a great extent on the legs, to a less degree on the arms and that it is about normal for a Filipino on the forehead. The apex beat in the mammary line, fifth intercostal space, right border of heart on the right line of sternum. Heart sounds normal, pulse 70. The man is somewhat constipated, but digestion and appetite are otherwise apparently normal. *Diagnosis:* beriberi.

On January 6, the man was put in the isolation room which we have described, and kept on a diet of rice, together with a quantity of fish. He was taught to collect his urine and fæces, to consume his food entirely, and the other details of the regimen necessary for experiments on metabolism. The diet to be given in the first set of experiments should be one which would *cause* beriberi; in the next period, organic phosphorus should be added; then a third period, like the first, should follow. In the next period after this, we intended to study the result of an increase of the intake of nitrogen alone without increasing the phosphorus, and in the last we desired to employ a diet more or less like that given to the first man, and which one of us has termed a typical Filipino diet, consisting of rice and fish. It was our further intention to double the two most important periods, namely, those with the diet poor and rich in phosphorus, so as to have a better control. Seven periods were therefore planned, each intended to cover four days, so that twenty-eight days would be necessary for the experiment.

The experiment originally began on January 9, but because of a failure in taking the fæces, we were compelled again to begin on January 10, so that the experiment covered twenty-nine instead of twenty-eight days. The arrangements were in all particulars exactly like those described above. The food used was again analyzed in respect to the rice, bacon, and fish; the bread and coffee were made in the same way as before and the values obtained in the first series were taken. As in the foregoing "normal" periods, we added bacon to the food of this patient so as to make it as deficient in phosphorus as possible. Three samples of bacon were employed: *A* was somewhat lean, containing some muscle fiber, whereas *B* and *C* consisted entirely of fat. The number of calories given, when reduced to the kilo of body weight (44 calories) was at least identical with the calories given to the first prisoners and therefore the food was quite sufficient to sustain a man who was not able to perform any muscular exertion. Phytin was added to the food during the two periods when a diet rich in phosphorus was administerd, so that we might study the result of the ingestion of this organic

phosphorus compound in beriberi. The phytin for this purpose was a commercial preparation, kindly delivered to us by the *Gesellschaft für Chemische Industrie*, Basel.

The egg albumen which was given during the period when the protein content of the food was increased was prepared by precipitating the whites of 100 eggs diluted by water, by means of sodium chloride and acetic acid at boiling temperature. The egg albumen was filtered, carefully washed and dried *in vacuo* at 60°. It was then finely powdered and the daily quantity cooked with the rice. Two preparations, termed egg albumen, I and II, showed only a very slight difference in their content of nitrogen. The fish used in the last period was canned salmon, carefully deprived of bones, the quantity needed for the entire experiment being taken at one time, mixed, and then divided into 4 portions of 100 grams each; a fifth portion of 100 grams being put aside and preserved with formalin as a sample for analysis. The samples of fish were kept on ice and, together with the rice, were cooked fresh each day. The rice, up to the last of January, was from the same stock, a white, fairly polished article. This was used up in February and the next supply, which is at present being used, shows a considerably higher content of phosphorus. This change in the supply of rice was disagreeable, but it could not be avoided.

During the first two periods, which we term A and B, the man was on a diet of bread, rice, bacon and sugar, similar to that given to the normal man in period V. He received about 44 calories, 0.18 gram of nitrogen and 0.026 gram P_2O_5 per kilo of body weight.

The result of this diet fully corresponds to that found with the normal man. It is another proof that a diet of bread and rice undoubtedly is not sufficient to fulfill the demand of the body for nitrogen and phosphorus. Therefore, these experiments clearly demonstrate that this same man, during the long time when he was kept on white bread before beriberi appeared, was constantly losing phosphorus and nitrogen.

Phytin was added during periods C and D. Despite the highly increased addition of phosphorus compounds, and in spite of the fact that this man should have a great demand for phosphorus because of his extended phosphorus starvation, he did not, like the normal man, retain phosphorus, but continued to lose this constituent, although in smaller amount than during the foregoing periods (A and B). The nitrogen balance also continued to be negative, almost to the same extent as in the foregoing periods.

Period E followed, in which a diet deficient in phosphorus again was given. During this time the loss in phosphorus in the urine as well as in the fæces was considerably less than during the corresponding periods (A and B), and therefore the relative lowering of the phosphorus balance was somewhat diminished. Possibly the diminution in the loss of phosphorus should be regarded as a result of the treatment with phytin. A marked reduction in the daily quantity of urine also took place, and this fact might also be taken in explanation; but, on the other hand, the reduction in the quantity of coffee may have been of influence.

The amount of nitrogen was increased during the next period (F) by the addition of egg albumen, in quantity so as to contain 8.34 grams instead of about 6.5 grams of nitrogen daily. In the normal man, the loss in nitrogen was reduced correspondingly by this addition, but in the individual suffering from beriberi, the same quantity of nitrogen as before was lost daily. It is still more remarkable that the excess of nitrogen did not appear almost entirely in the urine, but a large proportion was excreted in the fæces. Egg albumen, under normal conditions, should be absorbed almost to the extent of 98 per cent as was the case in period VI, but in this instance at least 25 per cent was lost.

The addition of 100 grams of fish, in period G, finally increased the nitrogen intake to 10.06 grams a day, or 0.25 gram per kilo of body weight. The nitrogen balance now registered only —0.6 gram, which did not represent full nitrogen equilibrium, although the shortage was small.

The phosphorus intake was at the same time increased to 1.91 grams per day, or 0.048 gram P_2O_5 per kilo of body weight. While this amount of P_2O_5 is far below that given during periods C and D, when phytin was added to the diet, the phosphorus balance now showed a daily loss of only 0.13 gram P_2O_5. Therefore, the man was nearly in phosphorus equilibrium. While during this period there was less P_2O_5 taken in, there still was a better retention of this element, probably because of the fact that, during this period, the loss of nitrogen was very small as compared with that in the foregoing periods C and D. It must be assumed that the destruction of the materials of the body containing nitrogen also brings with it the destruction of those containing phosphorus, and that therefore a loss in nitrogen should necessarily also be accompanied by one of phosphorus, regardless of the excess of phosphorus which may be taken in. Of course, as experiment VII has shown, we may find a favorable retention of phosphorus together with a loss of nitrogen, but in this instance the loss in phosphorus had taken place during a short period, whereas in the patient suffering with beriberi the loss of both nitrogen and phosphorus had continued for many weeks.

This view is strengthened by certain differences to be observed between cases of beriberi. A patient just beginning to show symptoms of nervous disorder and of weakness will often recover if his diet is changed in the course of one or two weeks; whereas, on the other hand, in instances where the disease has lasted several months, a long period of treatment is necessary before improvement sets in.

After finishing the metabolism experiment, we kept the patient on practically the same diet as he had received in period G after February 7, and, in addition, he received 4 grams of phytin daily, in two powders. He was maintained in isolation and under close observation. He was able to get up without help on or about February 20, toward the middle

of March he could stand with assistance, and also walk a few steps and now he is still weak, but free from clinical symptoms. He certainly has improved to a much greater extent than during the entire month of January, when there was practically no change in his condition. The dynamometric tests give objective data concerning his muscular and general condition.

Dynamometric tests.

Date.	Right.	Left.	Remarks.
January 9	55	35	
January 17	60	35-40	Jan. 18 to 25, 6 grams of phytin daily.
January 25	70	50	
January 30	65	45	
February 7	65	45	From Feb. 7, 4 grams of phytin daily.
February 15	75	50	
March 1	75	50	
March 7	75	55	

The most striking result of these metabolism experiments on a beriberi patient is that at the present stage of the disease he can not utilize the addition of phosphorus in phytin (periods C and D) or of protein (period F) *to the same extent as normal man* (periods IV, VI, and VII), and that he therefore requires a higher intake of nitrogen and phosphorus in similar food to reach nitrogen and phosphorus equilibrium (period G) than does a normal man (period III).

The results of our extended metabolism experiments under varying conditions, without doubt lead to the same conclusions as those of the Japanese investigators, namely, that nitrogen destruction in beriberi is *increased*. The objection that the man may have been undernourished can not be made, as was the case in Miura's experiment, because he took in a higher number of calories than both of the normal men on the same kind of food.

It is not our desire to decide from these experiments alone whether this higher demand is *characteristic* of beriberi or not, nor whether the greater demand for phosphorus and nitrogen is caused by some other reason and therefore predisposes the man to contract the disease, nor whether the higher demand is the result of the long phosphorus and nitrogen starvation which may have weakened the entire system. Further investigations along this line seem to be absolutely necessary.

The last period and the following dietary treatment show that an increased intake of phosphorus and nitrogen have a decidedly favorable influence on the patient. At the same time his nitrogen and phosphorus balance approaches equilibrium, while previously, during the time when there was no improvement, it was negative.

Some points worth mentioning still remain. The excretion of P_2O_5 as well as of nitrogen in period E, after phytin had been given, is considerably smaller than it was previously in periods A and B. The phytin (periods C and D) also increases the amount of total solids in the fæces, having a slight laxative action, without at the same time increasing the loss of nitrogen. The quantity of urine rises in the normal man, not only when phytin but also when *darac* is given; but there seems to have been no effect with the beriberi patient.

We do not believe the question of the effect of a constant phosphorus and nitrogen starvation to be fully solved by a study of the problem of beriberi. If our supposition is correct and if beriberi is caused by a lack of phosphorus and nitrogen in the diet, then there should be a great number of people whose diet is lacking in phosphorus and nitrogen. However, this starvation may not be of sufficient extent as to cause them to be actually ill. These individuals may be somewhat weak, less able to work and easily tired. The scale of transition between strong, healthy men, to those showing the typical paralysis of beriberi is probably extensive. We are fully convinced that the character of the food is not the least important cause of the inability to work which we notice in the lower classes of natives. The great problem of the influence of nourishment upon general health and muscular power is involved in this consideration.

CONCLUSIONS AND SUMMARY.

1. It is highly probable that living for an extended period on a one-sided almost exclusively vegetable diet, which is characterized by its poverty in phosphorus and protein, may result in beriberi.

2. The process of polishing rice removes a fine skin and the outer layers (bran); this rice bran is rich in phosphorus, especially in its organic, soluble form (phytin); the content of phosphorus of the rice is considerably reduced by the removal of the bran.

3. Polished rice, poor in phosphorus, may cause beriberi in man if it is the main constituent of the food; but it is harmless if sufficient other nourishment, rich in phosphorus and protein, is taken. The same polished rice causes a polyneuritis in chickens. White bread, a food of similar chemical composition as regards phosphorus and protein, can not sustain monkeys in normal health if it forms the entire diet.

4. The addition of phytin (the organic phosphorus compound from rice bran) considerably reduces the deleterious effect of white rice on chickens.

5 Metabolism experiments show that a diet such as is described in this paper, which contains about 40 calories per kilo, and which supplies less than 0.2 gram of nitrogen and 0.032 gram of P_2O_5 per kilo of body weight, can not meet the need of a normal man for phosphorus and protein. If phosphorus in the form of phytin or rice bran is added, a part

is stored and a favorable influence on the nitrogen metabolism can also be observed.

6. Metabolism experiments on a beriberi patient in a fairly advanced stage of the disease show that the capability of the man to utilize the nitrogen and phosphorus in the food is reduced; he demands a higher intake of nitrogen and phosphorus than a normal person to attain nitrogen and phosphorus equilibrium. It is especially to be noted that the capability of utilizing additional doses of phytin is considerably less than in that of a normal man under like conditions.

7. While it is certain that phosphorus and nitrogen starvation cause a certain and probably a great number of diseases which we term beriberi, there must be other factors,[44] especially when the œdematous form is observed.

RECOMMENDATIONS.

A very valuable portion of the rice is removed by the process of polishing and a healthy foodstuff transformed into one which is liable to cause a severe disease. Therefore, it should be the endeavor of all physicians to instruct rice producers and rice manufacturers concerning this process, and medical officers who have charge of the purchase of rice in large quantities for native troops, employees, etc., should use only grain which is either not polished or at least but slightly polished.[45]

Where the occurrence of beriberi in certain districts, in institutions, on ships, etc., is possible, care should be taken to give the people forr containing a sufficient quantity of phosphorus and protein. The greatest success would be attained by a large supply of fresh meat, or where this is not available, as in these islands, a sufficiency of the native bean, *mongo*.

A very promising means for the limitation or prevention of beriberi would be the use of the rice bran itself, 50 or 100 grams being cooked with the rice daily, as a protective or as a medicine. This rice bran seems to be the cheapest and most natural supply of the organic phosphorus compound, phytin; the preparation itself, while valuable, being much too expensive to be of any practical use. We are studying the value of rice bran as a protective on the ship *Pathfinder* of the United States Coast and Geodetic Survey, on which outbreaks of beriberi have periodically been recorded. Rice bran as a treatment has proved itself of value at Culion. The combined effect of a slightly increased protein intake with a constant supply of phytin showed favorable results in the treatment of the case in Bilibid, described above.

NOTE.—For the discussion on these papers see the end of this number.

[44] Nocht. *Arch. f. Schiffs-u. Trop.-Hyg. Beiheft.* (1908), 12, 5.

[45] These recommendations have already been fulfilled for the Philippine Islands by an executive order of His Excellency the Governor-General, by which "the use of polished rice in all public institutions is forbidden."

RECORDS OF THE EXPERIMENTS.

NORMAL MAN 1 (PERIODS III AND IV).

	Date.	Weight	Rice.	Bread.	Sugar.	Fish.	Coffee.	Darac (rice bran).	Carmine.	Remarks.
		Kilos.	*Grams.*	*Grams.*	*Grams.*	*Grams.*	*Cc.*	*Grams.*		
Period III.	Dec. 8	53.4	700	300	50		550			
	Dec. 9	54.3	600	300	50		300			
	Dec. 10		200	300	50	50	300		5 p. m.	
	Dec. 11	52.5	300A	300	50	[a]40	300			Fæces marked 4 p. m.
	Dec. 12	52.5	300A	300	50	[a]40	300			
	Dec. 13	52.5	300A	300	50	[a]40	300			
	Dec. 14	52.3	300A	300	50	[a]40	300		5 p. m.	
Period IV.	Dec. 15	52.3	250A	250	100	[a]40	300	75		Fæces marked 3 p. m.
	Dec. 16		250A	250	100	[a]40	300	75		
	Dec. 17		250A	250	100	[a]40	300	75		
	Dec. 18	52.5	250A	250	100	[a]40	300	75	5 p. m.	
	Dec. 19		ad libitum.							Fæces marked 9 a. m.

[a] Dried.

Analyses of foodstuffs.

	Nitrogen	P_2O_5.
	Per cent.	*Per cent.*
Fish[a] (dried)	6.30	0.70
Rice A	1.48	0.332
Bread, fresh	1.29	0.116
Coffee, 300 cc.	[b]0.16	[b]0.05

[a] 4.57 per cent fat. [b] Gram.

Analyses of urines.

Period.	Total quantity.	In 100 cubic centimeters.		Total.	
		Nitrogen.	P_2O_5.	Nitrogen.	P_2O_5.
	Cc.	*Grams.*	*Grams.*	*Grams.*	*Grams.*
III	3,992 (3,760)	0.798	0.1141	31.86	4.552
IV	4,545 (4,340)	0.632	0.1213	28.72	5.512

Analyses of fæces.

Period.	Total quantity air dry.	Solids.	Nitrogen.	P_2O_5.	Total quantity.		
					Solids.	Nitrogen.	P_2O_5.
	Grams.	*Per cent.*	*Per cent.*	*Per cent.*	*Grams.*	*Grams.*	*Grams.*
III	163.1	9.09	6.48	1.29	148.3	10.576	2.104
IV	298.0	8.70	5.02	4.51	259.2	14.96	13.84

Absorption.

	Nitrogen.		P_2O_5.	
	Period III.	Period IV.	Period III.	Period IV.
	Per cent.	*Per cent.*	*Per cent.*	*Per cent.*
Absorbed	75.94	67.45	68.5	36.6
Excreted in feces	24.06	32.55	31.5	63.4

PERIOD III.—*December 11 to 14, 1909.*

Food.	Nitrogen.	P_2O_5.	Calories (estimated).
	Grams.	*Grams.*	
200 grams sugar			800
1,200 grams bread	15.48	1.392	2,400
1,200 grams rice	17.76	3.984	4,000
160 grams fish, dried	10.08	1.120	320
1,200 cubic centimeters coffee	0.64	0.200	
Total intake	43.96	6.696	7,520
Per day	10.99	1.674	1,880

	Total.		Per day.			Balance.	
	Nitrogen.	P_2O_5.	Nitrogen.	P_2O_5.		Nitrogen.	P_2O_5.
	Grams.	*Grams.*	*Grams.*	*Grams.*		*Grams.*	*Grams.*
Outgo, urine	31.860	4.552	7.965	1.138	Intake per day	10.99	1.674
Outgo, feces	10.576	2.104	2.644	0.526	Outgo per day	10.61	1.664
Total	42.436	6.656	10.609	1.664	Balance	**+0.38**	**+0.010**

PERIOD IV.—*December 15 to 18, 1909.*

Food.	Nitrogen.	P_2O_5.	Calories (estimated).
	Grams.	*Grams.*	
400 grams sugar			1,600
1,000 grams bread	12.9	1.16	2,000
1,000 grams rice	14.8	3.32	3,300
160 grams fish, dried	10.08	1.12	320
1,200 cubic centimeters coffee	0.64	0.20	
300 grams darac	7.52	16.04	250
Total intake	45.94	21.84	7,570
Per day	11.49	5.46	1,890

	Total.		Per day.			Balance.	
	Nitrogen.	P_2O_5.	Nitrogen.	P_2O_5.		Nitrogen.	P_2O_5.
	Grams.	*Grams.*	*Grams.*	*Grams.*		*Grams.*	*Grams.*
Outgo, urine	28.72	5.512	7.18	1.378	Intake per day	11.49	5.46
Outgo, feces	14.96	13.840	3.74	3.460	Outgo per day	10.92	4.84
Total	43.68	19.352	10.92	4.838	Balance	**+0.57**	**+0.62**

NORMAL MAN 2 AND BERIBERI PATIENT (PERIODS V TO VIII AND A TO G).

Analyses of foodstuffs.

	Nitrogen.	P_2O_5.
	Per cent.	*Per cent.*
Rice I	1.32	0.28
Rice II	1.19	.57
Bread	1.29	.116
Bacon A	2.90	.215
Bacon B	.32	(.022)
Bacon C	.41	(.028)
Salmon	3.78	.42
Egg albumen I	13.71	
Egg albumen II	13.58	
Phytin		44.24
Capsules[a]	14.66	
Coffee, 500 cubic centimeters	[b].026	[b].016

[a] 10 capsules = 2.47 grams.
[b] Gram.

Normal Man 2.

Period and date.	Exper. Day.	Weight.	Rice.	Bread.	Sugar.	Coffee.	Bacon.	Egg albumin.	Capsules. Phytin.	Capsules. Carmine.	Urine. Total quantity.	Urine. Diluted to	Feces.
		K.	*G.*	*G.*	*G.*	*cc.*	*G.*	*G.*			*cc.*	*cc.*	
Jan. 25	1	64.8	ad libitum.							5 p. m.			
V Jan. 26	2	64.8	300 I	400	100	200	75 C				1,370	1,500	Marked off 10 a. m.
V Jan. 27	3	63.9	300 I	400	100	200	75 C				*740	1,000	
V Jan. 28	4	63.9	300 I	400	100	200	75 C				1,330	1,500	
V Jan. 29	5	63.7	300 I	400	100	200	75 C			4.30 p. m. 4 caps.	1,750	2,000	
VI Jan. 30	6	64.1	300 I	400	90	300	75 C	12 I			1,490	1,500	Marked off 10 a. m.
VI Jan. 31	7	63.7	300 I	400	90	300	75 C	12 I			2,310	2,500	
VI Feb. 1	8	63.7	300 II	400	90	300	75 C	12 II			1,495	1,500	
VI Feb. 2	9	64.1	300 II	400	90	300	75 C	12 II		5 p. m. 4 caps.	1,200	1,500	
VII Feb. 3	10	64.3	300 II	400	100	300	75 C		5 caps. 6 g.		2,200	2,500	Marked off 11 a. m.
VII Feb. 4	11		300 II	400	100	300	75 C		5 caps. 6 g.		2,370	2,500	
VII Feb. 5	12	63.9	300 II	400	100	500	75 B		5 caps. 6 g.		1,960	2,000	
VII Feb. 6	13	63.9	300 II	400	100	500	75 B		6 caps. 6 g.	4.30 p. m. 3 caps.	2,220	2,500	
Feb. 7	14		ad libitum.										Marked off 8 p. m.

[a] Dark.

Analyses of urines, V to VII.

Period.	Total quantity.	In 100 cubic centimeters.		Total.	
		Nitrogen.	P_2O_5.	Nitrogen.	P_2O_5.
	cc.	*Grams.*	*Grams.*	*Grams.*	*Grams.*
V	6,000 (5,190)	0.723	0.1157	43.38	6.94
VI	7,000 (6,495)	0.552	0.08956	38.64	6.269
VII	9,500 (8,750)	0.3476	0.06576	33.02	6.250

Analyses of fæces, V to VII.

Period.	Total quantity air dry.	Solids.	Nitrogen.	P_2O_5.	Total quantity.		
					Solids.	Nitrogen.	P_2O_5.
	Grams.	*Per cent.*	*Per cent.*	*Per cent.*	*Grams.*	*Grams.*	*Grams.*
V	209	89.42	6.49	1.40	186.9	13.56	2.93
VI	202	78.60	6.70	1.21	158.8	13.50	2.44
VII	192.3	89.42	6.73	3.84	172.0	12.94	7.38

PERIOD V.—*January 26 to 29, 1910.*

Food.	Nitrogen.	P_2O_5.	Calories (estimated).
	Grams.	*Grams.*	
1,200 grams rice	15.84	3.92	4,000
1,600 grams bread	20.64	1.86	3,200
300 grams bacon C	1.23	0.08	1,600
800 cubic centimeters coffee	0.44	0.13	
400 grams sugar			1,600
4 capsules	0.14		
Total intake	38.29	5.99	10,400
Per day	9.57	1.50	2,600

	Total.		Per day.			Balance.	
	Nitrogen.	P_2O_5.	Nitrogen.	P_2O_5.		Nitrogen.	P_2O_5.
	Grams.	*Grams.*	*Grams.*	*Grams.*		*Grams.*	*Grams.*
Outgo, urine	43.38	6.94	10.85	1.74	Intake per day	9.57	1.50
Outgo, fæces	13.56	2.93	3.39	0.73	Outgo per day	14.24	2.47
Total	56.94	9.87	14.24	2.47	Balance	**−4.67**	**−0.97**

PERIOD VI.—*January 30 to 31, February 1 to 2, 1910.*

Food.	Nitrogen.	P_2O_5.	Calories (estimated).
	Grams.	*Grams.*	
600 grams rice I	7.92	1.95	4,000
600 grams rice II	7.14	3.42	
1,600 grams bread	20.64	1.86	3,200
300 grams bacon C	1.23	0.08	1,600
24 grams egg albumen I	3.28		180
24 grams egg albumen II	3.26		
1,200 cubic centimeters coffee	0.62	0.20	
360 cubic centimeters sugar			1,440
4 capsules	0.15		
Total intake	44.24	7.51	10,420
Per day	11.06	1.88	2,605

	Total.		Per day.	
	Nitrogen.	P_2O_5.	Nitrogen.	P_2O_5.
	Grams.	*Grams.*	*Grams.*	*Grams.*
Outgo, urine	38.64	6.27	9.66	1.57
Outgo, feces	13.50	2.44	3.38	0.61
Total	52.14	8.71	13.04	2.18

	Balance.	
	Nitrogen.	P_2O_5.
	Grams.	*Grams.*
Intake per day	11.06	1.88
Outgo per day	13.04	2.18
Balance	−1.98	−0.30

PERIOD VII.—*February 3 to 6, 1910.*

Food.	Nitrogen.	P_2O_5.	Calories (estimated).
	Grams.	*Grams.*	
1,200 grams rice II	14.28	6.84	4,000
1,600 grams bread	20.64	1.86	3,200
150 grams bacon B	0.38	0.08	
150 grams bacon C	0.62	0.01	1,600
1,600 cubic centimeters coffee	0.83	0.26	
400 grams sugar			1,600
24 grams phytin		10.02	
24 capsules	0.90		
Total intake	37.65	19.65	10,400
Per day	9.41	4.91	2,600

	Total.		Per day.	
	Nitrogen.	P_2O_5.	Nitrogen.	P_2O_5.
	Grams.	*Grams.*	*Grams.*	*Grams.*
Outgo, urine	33.02	6.25	8.26	1.56
Outgo, feces	12.94	7.38	3.23	1.85
Total	45.96	13.63	11.49	3.41

	Balance.	
	Nitrogen.	P_2O_5.
	Grams.	*Grams.*
Intake per day	9.41	4.91
Outgo per day	11.49	3.41
Balance	−2.08	+1.50

Beriberi patient.

	Period and date.	Expr. Day.	Weight.	Rice.	Bread.	Sugar.	Coffee.	Bacon.	Capsules. Phytin.	Capsules. Carmine.	Urine. Total quantity.	Urine. Diluted to—	Fæces.
			K.	*G.*	*G.*	*G.*	*cc.*	*G.*			*cc.*	*cc.*	
	Jan. 9	1	40.8	200 I	300	100	500	30 A		5 p. m. 4 caps.	800	1,000	
A	Jan. 10	2	40.8	200 I	300	100	500	30 A			740	1,000	Marked off 8 a. m.
A	Jan. 11	3	40.8	200 I	300	100	500	30 A			1,490	1,500	
A	Jan. 12	4	40.8	200 I	300	100	500	30 A			2,940	3,000	
	Jan. 13	5	40.8	200 I	300	100	500	30 A			2,720	3,000	
B	Jan. 14	6	40.8	200 I	300	100	500	30 B			2,360	2,500	1 p. m. by regular course.
B	Jan. 15	7	40.6	200 I	300	100	500	30 B			1,100	1,500	
B	Jan. 16	8	40.8	200 I	300	100	500	30 B		5 p. m. 4 caps.	1,230	1,500	
	Jan. 17	9	40.4	200 I	300	100	500	30 B			1,690	2,000	
C	Jan. 18	10	40.4	200 I	300	100	500	30 B	5 caps. 6 g.		2,440	2,500	
C	Jan. 19	11	40.4	200 I	300	100	500	30 B	5 caps. 6 g.		2,075	2,500	3.30 p. m. marked off, not sharp.
C	Jan. 20	12	40.2	200 I	300	100	500	30 B	5 caps. 6 g.		2,900	3,000	
	Jan. 21	13	40.4	200 I	300	100	500	30 B	5 caps. 6 g.		2,375	2,500	
D	Jan. 22	14	40.4	200 I	300	100	500	30 B	5 caps. 6 g.		2,300	2,500	
D	Jan. 23	15	40.2	200 I	300	100	500	30 B	5 caps. 6 g.		3,000	3,000	4 p. m. by regular course.
D	Jan. 24	16	40.2	200 I	150	100	----	30 B	5 caps. 6 g.		1,900	2,000	
	Jan. 25	17	40.0	200 I	300	100	500	30 B	5 caps. 6 g.	4.30 p. m.	1,400	1,500	
E	Jan. 26	18	40.0	200 I	300	100	500	50 C			1,730	2,000	
E	Jan. 27	19	39.8	200 I	300	100	300	50 C			2,865	3,000	4 p. m. marked, not sharp.
E	Jan. 28	20	39.6	200 I	300	100	200	50 C			2,350	2,500	
	Jan. 29	21	40.0	200 I	300	100	----	50 C		5 p. m. 4 caps	[a]500	1,000	
F	Jan. 30	22	41.2	200 I	300	90	300	50 C	[b]12 g.		[a]445	1,000	
F	Jan. 31	23	41.2	200 I	300	90	300	50 C	[b]12 g.		1,170	1,500	10 a. m. marked off.
F	Feb. 1	24	40.8	200 II	300	90	----	50 C	[b]12 g.		1,785	2,000	
	Feb. 2	25	40.8	200 II	300	90	----	50 C	[c]12 g.	5 p. m. 4 caps.	1,205	1,500	
G	Feb. 3	26	40.8	200 II	300	100	----	----	[d]100 g.		960	1,000	
G	Feb. 4	27	----	200 II	300	100	----	----	[d]100 g.		1,575	2,000	7 p. m. marked off.
G	Feb. 5	28	41.0	200 II	300	100	----	----	[d]109 g.		1,230	1,500	
	Feb. 6	29	40.6	200 II	300	100	----	----	[d]100 g.	4.30 p.m. 3 caps.	2,840	3,000	
	Feb. 7	30	----	ad libitum.							995	1,000	
											----	----	11 a. m. marked off.

[a] Very dark. [b] Egg albumen I. [c] Egg albumen II. [d] Fish.

Analyses of urines, A to G.

Period.	Total quantity.	In 100 cubic centimeters.		Total.	
		Nitrogen.	P_2O_5.	Nitrogen.	P_2O_5.
	cc.	Grams.	Grams.	Grams.	Grams.
A	8,500 (7,890)	0.2872	0.0730	26.41	5.21
B	10,000 (8,820)	0.2154	0.0496	21.54	4.96
C	10,500 (9,650)	0.2094	0.050	21.99	5.25
D	8,500 (8,030)	0.272	0.05144	22.12	4.37
E 2 days	5,500 (5,215)	0.1995	0.03740	10.97	2.87
E 2 days	2,000 (945)	0.095		6.60	
F	6,000 (5,120)	0.8986	0.05604	28.92	3.36
G	7,500 (6,640)	0.3884	0.06036	29.13	4.53

Analyses of fæces, A to G.

Period.	Total quantity air dry.	Solids.	Nitrogen.	P_2O_5.	Total quantity.		
					Solids.	Nitrogen.	P_2O_5.
	Grams.	Per cent.	Per cent.	Per cent.	Grams.	Grams.	Grams.
A	164.0	94.54	8.12	1.78	155.1	13.32	2.92
B	174.5	96.44	7.90	1.93	168.3	13.79	3.37
C	200.0	96.24	6.77	5.77	192.5	13.54	11.54
D	234.7	89.15	6.39	5.27	208.6	15.00	12.36
E	195.6	91.48	7.58	1.34	178.9	14.83	2.62
F	189.7	86.63	9.38	1.67	164.3	17.79	3.16
G	156.0	93.92	8.60	2.34	146.5	13.42	3.65

PERIOD A.—*January 10 to 13, 1910.*

Food.	Nitrogen.	P_2O_5.	Calories (estimated).
	Grams.	Grams.	
800 grams rice I	10.56	2.44	2,650
1,200 grams bread	15.48	1.39	2,400
120 grams bacon A	3.48	0.26	600
2,000 cubic centimeters coffee	1.04	0.32	
400 grams sugar			1,000
Total intake	30.56	4.41	7,250
Per day	7.64	1.10	1,810

	Total.		Per day.			Balance.	
	Nitrogen.	P_2O_5.	Nitrogen.	P_2O_5.		Nitrogen.	P_2O_5.
	Grams.	Grams.	Grams.	Grams.		Grams.	Grams.
Outgo, urine	26.41	6.21	6.60	1.55	Intake per day	7.64	1.10
Outgo, fæces	13.32	2.92	3.33	0.73	Outgo per day	9.93	2.28
Total	39.73	9.13	9.93	2.28	Balance	−2.29	−1.18

PERIOD B.—*January 14 to 17, 1910.*

Food.	Nitrogen.	P_2O_5.	Calories (estimated.)
	Grams.	*Grams.*	
800 grams rice I	10.56	2.44	2,650
1,200 grams bread	15.48	1.39	2,400
120 grams bacon B	0.38	0.03	600
2,000 cubic centimeters coffee	1.04	0.32	
400 grams sugar			1,600
4 capsules	0.15		
Total intake	27.61	4.18	7,250
Per day	6.90	1.05	1,810

	Total.		Per day.			Balance.	
	Nitrogen.	P_2O_5.	Nitrogen.	P_2O_5.		Nitrogen.	P_2O_5.
	Grams.	*Grams.*	*Grams.*	*Grams.*		*Grams.*	*Grams.*
Outgo, urine	21.54	4.96	5.39	1.24	Intake per day	9.90	1.05
Outgo, fæces	13.79	3.37	3.45	0.84	Outgo per day	8.84	2.08
Total	35.33	8.33	8.84	2.08	Balance	−1.94	−1.03

PERIOD C.—*January 18 to 21, 1910.*

Food.	Nitrogen.	P_2O_5.	Calories (estimated).
	Grams.	*Grams.*	
800 grams rice I	10.56	2.44	2,650
1,200 grams bread	15.48	1.39	2,400
120 grams bacon B	0.38	0.03	600
2,000 cubic centimeters coffee	1.04	0.32	
400 grams sugar			1,600
24 grams phytin		10.62	
20 capsules	0.75		
Total intake	28.21	14.80	7,250
Per day	7.05	3.70	1,810

	Total.		Per day.			Balance.	
	Nitrogen.	P_2O_5.	Nitrogen.	P_2O_5.		Nitrogen.	P_2O_5.
	Grams.	*Grams.*	*Grams.*	*Grams.*		*Grams.*	*Grams.*
Outgo, urine	21.99	5.25	5.50	1.31	Intake per day	7.05	3.70
Outgo, fæces	13.54	11.54	3.36	2.89	Outgo per day	8.86	4.20
Total	35.53	16.79	8.86	4.20	Balance	−1.81	−0.50

PERIOD D.—*January 22 to 25, 1910.*

Food.	Nitrogen.	P_2O_5.	Calories (estimated).
	Grams.	*Grams.*	
800 grams rice I	10.56	2.44	2,650
1,050 grams bread	13.55	1.22	2,100
120 grams bacon B	0.38	0.03	600
1,500 cubic centimeters coffee	0.78	0.24	
400 grams sugar			1,600
24 grams phytin		10.62	
20 capsules	0.75		
Total intake	26.02	14.55	6,950
Per day	6.51	3.64	1,740

	Total.		Per day.			Balance.	
	Nitrogen.	P_2O_5.	Nitrogen.	P_2O_5.		Nitrogen.	P_2O_5.
	Grams.	*Grams.*	*Grams.*	*Grams.*		*Grams.*	*Grams.*
Outgo, urine	22.12	4.37	5.53	1.09	Intake per day	6.51	3.64
Outgo, fæces	15.00	12.36	3.75	3.09	Outgo per day	9.28	4.18
Total	37.12	16.73	9.28	4.18	Balance	−2.77	−0.54

PERIOD E.—*January 26 to 29, 1910.*

Food.	Nitrogen.	P_2O_5.	Calories (estimated).
	Grams.	*Grams.*	
800 grams rice I	10.56	2.44	2,650
1,200 grams bread	15.48	1.39	2,400
1,000 cubic centimeters coffee	0.52	0.16	
200 grams bacon C	0.82	0.06	1,000
400 grams sugar			1,600
4 capsules	0.15		
Total intake	27.53	4.05	7,650
Per day	6.88	1.01	1,910

	Total.		Per day.			Balance.	
	Nitrogen.	P_2O_5.	Nitrogen.	P_2O_5.		Nitrogen.	P_2O_5.
	Grams.	*Grams.*	*Grams.*	*Grams.*		*Grams.*	*Grams.*
Outgo, urine (2 days)	10.97	2.81	4.39	0.70	Intake per day	6.88	1.01
Outgo, urine (2 days)	6.60						
Outgo, fæces	14.83	2.62	3.71	0.66	Outgo per day	8.10	1.36
Total	32.40	5.43	8.10	1.36	Balance	−1.22	−0.35

PERIOD F.—*January 30 to February 2, 1910.*

Food.	Nitrogen.	P_2O_5.	Calories (estimated).
	Grams.	*Grams.*	
400 grams rice I	5.28	1.22	2,650
400 grams rice II	4.76	2.28	
1,200 grams bread	15.48	1.39	2,400
200 grams bacon C	0.82	0.06	1,000
600 cubic centimeters coffee	0.81	0.10	
360 grams sugar			1,440
36 grams egg albumen I	4.94		180
12 grams egg albumen II	1.63		
4 capsules	0.15		
Total intake	33.37	5.05	7,670
Per day	8.34	1.26	1,920

	Total.		Per day.			Balance.	
	Nitrogen.	P_2O_5.	Nitrogen.	P_2O_5.		Nitrogen.	P_2O_5.
	Grams.	*Grams.*	*Grams.*	*Grams.*		*Grams.*	*Grams.*
Outgo, urine	23.92	3.36	5.98	0.84	Intake per day	8.34	1.26
Outgo, feces	17.79	3.16	4.45	0.79	Outgo per day	10.43	1.63
Total	41.71	6.52	10.43	1.63	Balance	**−2.09**	**−0.37**

PERIOD G.—*February 3 to 5, 1910.*

Food.	Nitrogen.	P_2O_5.	Calories (estimated).
	Grams.	*Grams.*	
800 grams rice II	9.52	4.56	2,650
1,200 grams bread	15.48	1.39	2,400
400 grams sugar			1,600
400 grams fish (salmon)	15.12	1.68	500
3 capsules	0.12		
Total intake	40.24	7.63	7,150
Per day	10.06	1.91	1,790

	Total.		Per day.			Balance.	
	Nitrogen.	P_2O_5.	Nitrogen.	P_2O_5.		Nitrogen.	P_2O_5.
	Grams.	*Grams.*	*Grams.*	*Grams.*		*Grams.*	*Grams.*
Outgo, urine	29.13	4.58	7.28	1.13	Intake per day	10.06	1.91
Outgo, feces	13.42	3.65	3.36	0.91	Outgo per day	10.64	2.04
Total	42.55	8.18	10.64	2.04	Balance	**0.58**	**−0.13**

SOME OBSERVATIONS CONCERNING BERIBERI.[1]

By GOROSAKU SHIBAYAMA.[2]

(From the Institute for Infectious Diseases, Tokio.)

Marked attention has been given in Japan to the question of the etiology of beriberi since the Russo-Japanese war, not only in medical circles, but also in a practical manner. During the war, Japan had 200,000 cases of beriberi in the army, and this was the only ravaging epidemic with which the army had to contend. A beriberi commission, consisting of bacteriologists, pathologists, chemists and clinicians was therefore formed two years ago by the ministry of war, and I was one of the commission, who, together with two colleagues, made a journey to the Netherlands East Indies in order to observe the occurrence of beriberi in that region.

Beriberi is now very widely distributed in Java and Sumatra, much more so than in former years. The disease is present among a number of Chinese on the Island of Banka, where the tin mines are situated. We made a number of observations in this locality, which I wish briefly to mention in this place.

The present views concerning the etiology of beriberi are very diverse, and this is not the place to enter more extensively into a discussion of the literature, but the theory of the relation of the disease to the consumption of rice must here be considered. Ten years ago Eykman conducted some experiments with fowls, producing polyneuritis by the exclusive feeding of husked (polished) rice, whereas the unhusked (red) variety did not produce this result. Vordermann then demonstrated, in conformity with this work, that beriberi is a much rarer disease in prisons in Java where unhusked rice is used than in institutions in which the polished grain is employed. However, I wish to caution against regarding polyneuritis of fowls as being identical with human beriberi, and the observations of Vordermann leave many lapses and have much to be brought against them. Beriberi, as a fact, is endemic among oriental peoples who also use rice as the chief article of diet,

[1] Read at the first biennial meeting of the Far Eastern Association of Tropical Medicine, at Manila, March 11, 1910. Translated from the German by P. C. F.

[2] Delegate from His Imperial Japanese Majesty's Government.

and therefore the theory of the connection of the disease with the consumption of rice is always brought to the fore. The observations of Braddon, Fletcher, Ellis and Fraser, in the Malay Peninsula, have again called attention to this matter. According to the observations of these English authors, the consumption of parboiled rice is able to prevent the outbreaks of beriberi in hospitals and the disease can even be cured by the ingestion of this variety.

Upon the basis of the theory of the connection of rice with the etiology of beriberi, a freshly husked, but not polished, rice was used in some of the mines on Banka Island, but the result was in evident contradiction to the theory.

In 1908, 1,195 cases of beriberi developed in the Blinjoe, one of the mining districts. Mine No. 3 was especially unfortunate, for 166 out of 410 workmen contracted the disease, and mine No. 4 developed 118 cases among 390. No. 5 had 400 workmen, and 97 of these were ill with beriberi; on the other hand, the remaining mines showed but few cases. For two years the workmen had received unpolished, fresh rice, not only in mines Nos. 3, 4, and 5, but also in No. 11, in which latter 49 out of 300 workmen contracted the disease; on the other hand, the laborers in the remainder always had polished and old rice. It may further be stated, according to Hullshoff-Pol, that the workmen in all the mines received 150 grams *kadjang idjo* beans, together with dried fish and fresh vegetables, daily. The result of our observations, therefore, was as follows:

1. Even if the workmen in the mines receive 150 grams of *kadjang idjo* regularly every day, nevertheless beriberi occurs among them.

2. Even if the laborers are given a diet of fresh, unpolished rice, nevertheless they develop more cases of beriberi than those in the other mines, where they receive polished and older Java rice.

I therefore could not find the assumption to be confirmed that unpolished rice, which has the same composition as parboiled rice, could prevent beriberi.

I wish to add some statements concerning a general epidemic on board a steamship which took 600 emigrants from Yokohama to South America; and 62 of whom were for various causes returned to that port with the same vessel. The entire voyage out and home took one hundred and thirty-nine days. Beriberi gradually appeared among the returning emigrants and by the time they reached Yokohama all, without a single exception, had contracted the disease, and 6 died. The remainder, upon arrival in port, were transferred to a hospital, where I saw the patients, and after a careful examination I proved that the sickness was genuine beriberi.

The question has not been decided as to whether the general epidemics which at times occur on board ship and which so greatly resemble beriberi are in reality always genuine beriberi, or only a similar condition or a scorbutic disease brought about through lack of nourishment. The Norwegian commission, in expressing an opinion concerning the beriberi

of sailing ships, believed the disease to be genuine beriberi, whereas other authors, for example, Nocht in Hamburg, maintain that the condition is dependent on defect in nourishment. In my case, at least, the disease was genuine beriberi.

Furthermore, in the past summer, I observed an epidemic of beriberi in certain fishing villages of Japan.

All my observations lead me to the conclusion that uniform, but little changing, monotonous diet predisposes to the disease. The condition of nourishment of the Chinese in two of the mines of Banka was fairly good, the total quantity of the chief constituents of diet, namely, protein, fat and carbohydrates, was sufficient, but the diet was always one-sided and not varied throughout the year. This is also true of the general epidemic of beriberi aboard the steamship referred to above, and in the fishing villages the one-sided diet was the only point to be observed.

However, the one-sided or monotonous diet is only the predisposing cause of beriberi; the true cause must be sought in other directions. This can be illustrated by an example. Abdominal typhoid is treated in Europe and America, as well as in Java, or for that matter in the entire Orient, by means of a liquid diet, especially milk. In the Orient, beriberi very frequently occurs as a complication among the convalescent patients; whereas this has never been observed in Europe or America. It is also true in the Orient that during sieges of cities and under other circumstances where there is insufficiency of diet, beriberi is very frequently observed, whereas this is not the case in the Occident. It is therefore not unreasonable to assume that the microörganisms of beriberi are only present in the Orient and, given a predisposing cause, are capable of causing the disease, whereas in the West beriberi does not appear, owing to the absence of the infecting organisms, although the same favorable predisposing cause may be present.

FOOD SALTS IN RELATION TO BERIBERI.[1]

By E. D. KILBOURNE.[2]

Beriberi has probably existed in the Philippine Islands since very early times. Schneider[3] reported cases at Zamboanga in 1852. Koeniger[4] is, I believe, in error, in stating that beriberi did not exist among the natives prior to 1882, although it was probably less common than it is now. During and since the year 1877 the imports of rice have greatly exceeded the exports. From our present knowledge of the etiologic relation of polished rice to beriberi, it is safe to say that the disease was not absent during these and during previous years when the imports were not inconsiderable.

TABLE I.—*Imports and exports of rice into Manila, in kilos.*

Year.	Imported.	Exported.	Year.	Imported.	Exported.
1851–1857	None.	Not known.	1884	108,431,626	1,609
1858	1,393,040	772,920	1885	42,440,640	749
1859	746,460	820,440	1886	61,798,722	18,585
1860	1,758,240	6,313,428	1887	79,987,973	82,337
1861	16,218	2,443,500	1888	82,445,441	374
1862–63	Not known.	Not known.	1889	85,417,158	905
1864	1,046,100	20,396,300	1890	71,166,714	13,426
1865	258,257	21,650,046	1891	72,664,363	None.
1866	4,788	6,000,054	1892	62,709,197	254
1867	480,804	2,036,696	1893	41,000,503	62,492
1868–1872	Not known.	Not known.	1894	44,870,685	1,513,658
1873	7,311,002	18,278	1895–1897[a]	Not known.	Not known.
1874	10,811,589	101,371,178	1898–99[b]	55,817,073	None.
1875	2,910,847	1,524,855	1899–1900	110,141,537	None.
1876	239,539	3,571,931	1900–1901	178,605,867	None
1877	23,005,946	86,800	1901–2	216,812,362	None.
1878	23,670,099	265,285	1902–3	307,835,856	None.
1879	58,818,165	89,848	1903–4	330,518,006	None.
1880	13,555,647	366,241	1904–5	256,037,430	None.
1881	5,558,047	203,977	1905–6	138,341,469	None.
1882	8,971,305	840	1906–7	112,985,635	None.
1883	54,414,683	50,361	1907–8	162,515,826	None.

[a] No Spanish records could be found at the custom-house later than 1894.

[b] The American records begin with August 20, 1898, and show the amounts in the fiscal years ending June 30.

[1] Read at the first biennial meeting of the Far Eastern Association of Tropical Medicine, in Manila, March 11, 1910.

[2] Captain, Medical Corps, United States Army, Member of the United States' Army Board for the Study of Tropical Diseases as They Exist in the Philippine Islands.

[3] *Geneesk. Tijd. v. Ned. Ind.* (1890), **30**, 438.

[4] *Deutsches Arch. f. klin. Med.* (1884), 34, 419.

Beriberi has been endemic among the native population since the occupation of the Philippines by the American forces in 1898, and several cases have been reported among the whites. Although I have examined several white soldiers and civilians, said to have beriberi, I can not say that I have ever seen a genuine case in this class, and am of the opinion that the disease is extremely rare among them. In the Tropics it is easy to confuse multiple neuritis from other causes with true beriberi.

The relationship of rice to beriberi is so well established that a consideration of one must include the other; therefore, a brief outline of the rice-eating habits of the inhabitants of the Islands is here given. Nearly all the tribes of the Philippines are of the Malay race, and though not all are of the same religious belief, their modes of preparing rice for eating are practically identical. None, as far as I can ascertain, use parboiled rice. There are a considerable number of Chinese and a few East Indians all of whom use polished rice, either native or imported. The latter prefer an Indian polished rice, which they import in small amounts for their own consumption.

The inhabitants of the interior use irrigated rice, when practicable, and mountain or upland rice when water is not accessible. Those in the lower lands, for the greater part, consume irrigated native rice. The inhabitants of the coastal plains use the latter, or imported rice brought, principally, from the British and French East Indies and from Siam.

For twenty-one years preceding, and since the American occupation, there has not been enough native rice produced to supply the population. Before 1877 this was not so; then the rice exported exceeded that imported. The various sources and amounts of rice imported since the American occupation are given in the following table, compiled from the records of the Bureau of Customs.

TABLE II.—*Sources of imported rice—fiscal years ending June 30—weights in kilos.*

Year.	United States.	United Kingdom.	France.	Spain.	Japan.	British East Indies.	Dutch East Indies.	French East Indies.	Siam.	Other Asia.
1899	.....	.....	.....	859,272	5,727	3,172,052	.....	6,774,902	.....	.....
1900	.....	.....	.....	6,619	129	5,589,964	.....	149,999,369	.....	.....
1901	.....	505	82	136,405	147	4,624,958	7,500	161,647,368	12,188,900	.....
1902	218,298	.....	.....	16,713	85,421	22,819,860	1,919	179,719,883	13,414,030	700,780
1903	6,665	.....	.....	42,040	8,258	27,687,089	.....	262,930,276	16,481,171	630,354
1904	.....	.....	.....	30,859	.....	38,443,895	29,547	264,317,216	(a)	26,519,980
1905	.....	.....	100	19,381	7,287	18,432,723	198,535	208,772,632	(a)	28,606,778
1906	.....	.....	.....	25,872	9,837	8,412,494	1,376	120,312,056	(a)	9,988,451
1907	.....	.....	.....	17,901	11,616	971,600	.....	106,673,410	5,311,105	.....
1908	.....	.....	.....	35,773	31,398	1,209,207	2,739	152,799,236	8,427,425	.....

[a] Siam rice 1904–1906 is included in that from "Other Asia."

The rice used in the Philippines, as regards preparation, is of three kinds: That grown at home and pounded out by hand in large wooden mortars; that grown at home and submitted to milling of varying degrees of thoroughness; and the imported rice, all of which has been milled and polished abroad.

The first variety, home grown and hand produced, is used in the rural districts and by about one-half of the natives in the coast towns. The second and third varieties, polished native and imported rice, are used principally in the coast towns and their immediate vicinity. There are regions where little or no rice is consumed, as in the Batan Islands, just north of Luzon. Meat and vegetables constitute the diet of the inhabitants of these islands; they have no beriberi. In the town of Aparri, 120 miles south of the Batan group, imported polished rice is found for sale in the *tiendas* and beriberi exists among the natives. On the contrary, in the coast towns of Casiguran and Baler, which are around the northeastern corner of Luzon from Aparri, no imported rice was observed in the *tiendas* and no beriberi among the people.

In general, beriberi is found in the coast towns and along the lines of communication, and in the same places polished rice is extensively eaten. At Taytay, in Rizal Province, a small town of 6,000 inhabitants about 15 miles from Manila, beriberi is remarkably infrequent, although the town is on the railroad. As a matter of fact, however, polished rice seems to be used there little, if any. Samples of rice gathered at random in the *tiendas* of Taytay were all of the home-grown varieties and incompletely milled.

Our observations seem to prove that beriberi in the Philippines is found only where polished rice, either imported or home grown, is used, principally along the coasts or rivers. No doubt the new railway system will open up increased areas where polished rice will be consumed and the distribution of beriberi in the Philippine Islands may be changed in the next few years.

This etiologic relation of polished rice to beriberi agrees with the findings of other workers. If polished rice is a factor in the causation of beriberi, and this seems to us to be proved, it must be because it either conveys into the body some extraneous poisonous substance, or else is deficient in, or, at least, fails to give to the body enough of some needed element or elements. The work done in the Philippines by Major Dutcher and others, and our own observations, have failed to give any support to the first idea, consequently our researches have been directed to an investigation of the privation theory.

It is not sufficient to make deductions from analyses of the various rices, polished and parboiled, or to draw conclusions from experiments in which such rices are fed to lower animals, although such experiments may be suggestive and point out a profitable line of research in the human. The ingestion by the human of other articles of diet, rich in a certain element, may entirely compensate for the lack of such element in the rice eaten, so unless the entire diet of a body of men for a period equal, at least, to the so-called "period of incubation" is known, no

reliable conclusions can be drawn. From the analysis of one predominating article of food we might conclude that the Irish peasant has this or that disease because the potatoes, which constitute a large part of his diet, contain a very small percentage of phosphorus. If we examine his entire ration we find that this element is amply supplied by milk and eggs, which are easily and cheaply obtained. Eykman [4] and Sakaki [5] have shown that fowls fed on polished rice develop polyneuritis in five to ten weeks, but horses develop a condition akin to beriberi [6] when fed on unhusked grain, the food which prevents the condition in the case of the fowls. We have no means of knowing that these diseases in animals are caused by the same conditions that induce human beriberi. Monkeys, in our laboratory, fed on boiled, polished rice and water alone, showed no bad effects for fourteen weeks and actually gained in weight.

To determine in what respect the diets containing a considerable proportion of polished rice are at fault, the United States Army Board for the Study of Tropical Diseases as They Exist in the Philippines, of which I am a member, has, during the past year, studied the situation as it exists in the native troops (Philippine Scouts). To obtain reliable and accurate data from the civilian natives is impossible; only in a military organization, prison, school, or other similar body is it possible to exercise a control sufficient to make the findings of any value. Consequently, our observations on the civilian population are of a general nature only and made from material incidentally gathered while working on beriberi among the Scouts.

The Scouts were organized in 1901. The annual reports of the Surgeon-General of the United States Army first mention beriberi as occurring among individuals of this body of troops in 1902.

Beriberi in the Philippine Scouts.

Year.	Admissions per 1,000.	Deaths per 1,000.	Year.	Admissions per 1,000.	Deaths. per 1,000
1902	7.75	0.38	1906	36.98	1.79
1903	9.42	.87	1907	24.58	1.28
1904	74.62	1.52	1908	121.53	1.35
1905	35.93	1.21			

In 1909 the admissions of scouts to sick report, because of beriberi, were only exceeded in number by two other causes, malaria and miscellaneous digestive troubles.

It was evident, after a study of the data obtained from the records of these companies, that the amounts of proteid, fat and carbohydrate actually consumed were sufficient.

[4] Polyneuritis bij Hoenders. *Geneesk. Tijd. v. Ned. Ind.* (1896), 36.
[5] *Sai-i-Kwai* (1903), March 31, April 30.
[6] Braddon, W. L. The Cause and Prevention of Beriberi. (1907), 350.

The Filipino ration (issued to the Scouts) is as follows:

Component articles.	Quantities.	Substitutive articles.	Quantities.
	Ounces.		*Ounces.*
Beef, fresh	12	Bacon	8
		Canned meat	8
		Fish, canned	12
		Fish, fresh	12
Flour	8	Hard bread	8
Baking powder (in field)	0.32		
Rice	20		
Potatoes	8	Onions	8
Coffee, roasted and ground	1		
Sugar	2		
Vinegar	*0.08		
Salt	0.64		
Pepper	0.02		

*Gill.

A considerable latitude is allowed in the amounts consumed. For example: If the company does not care to use a certain article of food on the ration list, it need not draw it from the commissary, but will in its place receive a commuted value, in cash, at the end of the month, with which to purchase other articles from outside sources. Also, the company may have a substantial "company fund" derived from its share of the profits of the post exchange and post bakery with which to buy desired articles of food to enrich the mess. When the company is stationed in an inaccessible place, away from markets, it must necessarily live almost entirely from the commissary; but when quartered near good markets it may be able to buy fresh vegetables and other things cheaply and draw little from the commissary. In these ways the ration actually eaten by the men may differ considerably from that laid down by the Army Regulations.

To determine exactly what the dietaries were, the company and subsistence department records were consulted and the actual articles and amounts ascertained. In no case were the amounts of proteid, fat and carbohydrate found deficient. The Filipino ration, as listed, contains approximately the following: Proteid, 122 grams; fat, 45 grams; carbohydrates, 685 grams. The proteid and fat are ample, and the amount of carbohydrates is more than enough for a Filipino who weighs about four-fifths as much as the average American.

In recent years the rôle played by the inorganic salts in the human economy has been given a great deal of study. In the last decade much work has been done by physiologists on the action of salts on the healthy organism, but the relation of the different salts to known pathologic conditions has only partially been worked out.

The principal salts introduced with the food are combinations of

sodium, potassium, calcium, and magnesium, with carbonic, hydrochloric, sulphuric, and phosphoric acids. If a deficiency in any of these has an etiologic bearing on beriberi, such deficiency should appear both in the polished rice and in the diets of the "infected" companies.

A comparative table, complied from data taken from Blyth's "Foods, Their Composition and Analysis," is as follows:

TABLE III.—*Comparative table of salts in food (Blyth).*

Constituents.	Rice.	Potatoes.	Peas.	Corn.
Potash	0.0977	0.5050	1.1393	0.2390
Soda	.0247	.0215	.0254	.0608
Lime	.0145	.0210	.1322	.0352
Magnesia	.0504	.0354	.2109	.1232
Ferric oxide	.0055	.0096	.0227	.0135
Phosphoric acid	.2415	.1421	.9653	.5904
Sulphuric acid	.0027	.0532	.0956	.0068
Silica	.0121	.0174	.0227	.0301
Chlorine	.0004	.0255	.0408	.0011

It will be noted that rice is low in potash, lime, phosphoric acid, sulphuric acid and chlorine. The potassium, calcium, and phosphorus content will be discussed later. The low sulphuric acid content is of no importance.

As has been stated above, the amounts of proteid have been sufficient in the dietaries of all the companies investigated, thereby furnishing the amount of sulphur necessary. "Sulphates eaten pass out through the urine. They play no part in the life of the cell."[7] The low chlorine content has no bearing on beriberi among the Scouts, for the native soldier consumes enough table salt to make up the deficiency; whether he eats too much salt and thereby extracts needed potassium ions from the body is a question worthy of consideration.

The phosphorus and potassium content of the dietaries of several Scout companies and of the prisoners at work on Corregidor Island is given in the following table. The companies have been divided into three classes according to the incidence of beriberi in each.

Average number of grams consumed per man per day.

Class.	P_2O_5.	KCl.
I. Having many cases (5 organizations)	3.3474	1.0600
II. Having a few scattered cases (7 organizations)	3.9399	1.1905
III. Having no cases (3 organizations)	4.6279	1.6517

[7] American Text-Book of Physiology. Philadelphia, 1 ed. (1896), 951.

The amounts of phosphorus were computed from percentages given by Balland[8] and the amounts of potassium from Garrod's table.[9]

The amounts of P_2O_5 average 3.3474 grams in the bad companies, 3.9399 in those slightly affected, and 4.6279 in the companies and the prisoners having no beriberi; a difference of 1.2805 grams between the worst and the best averages.

Just what amount of phosphorus is necessary for man, I am unable to say. About 3.5 grams phosphoric acid are excreted daily in the urine, and some phosphates of the alkaline earths in the fæces, but even though we were to determine exactly the intake and outgo we should still be unable to ascertain the amount required, for some of the phosphorus eliminated in the fæces may have been absorbed, used, and returned again for expulsion. A better idea of the amount required, as far as beriberi is concerned, may be gained from the phosphorus content of diets known to prevent beriberi. Such a diet is the garrison ration which is issued to our white soldiers. It contains 6.3433 grams P_2O_5. Four-fifths of this amount, to correspond to the weight of a Filipino, would be 5.0746 grams. It would seem that this is more than enough. In our third group, consuming an average of 4.6729 grams P_2O_5 daily, no beriberi occurred. The Filipino ration, as listed, contains 4.1768 grams P_2O_5.

The phosphorus of the food enters the body in three general classes, the soluble phosphates of sodium and potassium, the slightly soluble calcium and magnesium phosphates, and in organic combinations with nuclein, casein, and caseoses. It is not known whether or not calcium phosphate is absorbed from the intestinal tract. It is probable that the phosphoric acid ions are absorbed, chiefly, in combination with sodium and potassium.[10] From the feeding experiments of Hart, McCollum and Humphry,[11] it would appear that the phosphorus and potassium of the food are closely associated in their relation to the nutritive processes of the body.

The amounts of potassium chloride average 1.0600 grams in the worst, 1.1905 in the medium, and 1.6517 grams in the best organizations, a difference of 0.5917 grams between the worst and the best. These amounts of potassium chloride do not run exactly parallel to the phosphorus content of the three groups, there being a greater relative difference in the amounts of the former. The increase of the best over the worst was 56.76 per cent in potassium chloride, but only 38.22 per cent in P_2O_5.

The amount of potassium required daily by man is variously estimated, but is usually given as from 3 to 4 grams. While 3 grams of potassium

[8] *Bull. Acad. Med.* (1906), **56**, 612.
[9] Nothnagel. Encyclopedia of Practical Medicine.
[10] Herter. Chemical Pathology, 115.
[11] *Am. Journ. Phys.* (1909), **23**, 246.

chloride are found in the garrison ration, there are only 1.3 in that of the Filipino troops, a remarkable difference. An average of 1.6517 grams was found in our third or beriberi-free group.

An important influence of the potassium salts has been noted by Bunge.[12] "If a potassium salt be in solution together with sodium chloride, the two partially react on each other, with formation of potassium chloride. If now potassium carbonate, for example, be eaten, the same reaction occurs in the body: $K_2CO_3 + 2NaCl \lesssim 2KCl + Na_2CO_3$. The kidney has the power of removing soluble substances which do not belong to the blood, or are present in it to excess, and consequently the two salts formed as above are excreted. Hence, potassium carbonate has caused a direct loss of sodium and chlorine. For this reason, if potatoes and vegetables rich in potassium salts are eaten, sodium chloride must be added to the food to compensate for the loss. Natives living on rice do not need salt, for here the potassium content is low. Tribes living solely on meat or fish do not use salt, but care is taken that the animals slaughtered for food shall not lose the blood rich in sodium salts, and strips of meat dipped in blood are, by some races, considered a delicacy." If this be true, an excess of sodium salts will cause a loss of potassium in the same manner.

Jaques Loeb[13] has shown the necessity for a balance between the Na, Ca, and K ions, and that the Ca and K ions counteract the effects of the Na ions in the blood. When marine animals were placed in a pure solution of sodium chloride of the same concentration as sea water, their muscular contractility was lost. Small amounts of Ca and K ions antagonized the poisonous effects of the Na ions.

Herter gives the daily amount of sodium chloride necessary for a man as about 8 grams, which would be about 6.5 grams for a Filipino. The average daily amount of table salt consumed by each man in the organizations we investigated was 10 grams. This amount, ordinarily not harmful, since most people consume quantities far in excess of the physiological requirements, might be too much for diets as low in K ions as are those of the Scouts.

The amounts of calcium ingested were greatly in excess of their physiological needs.

The necessity of a proper balance in food salts is shown by the relation of varying amounts of sodium chloride to the œdema of nephritis, and of calcium chloride to epilepsy and rickets.

A deficiency in the amounts of potassium and phosphorus, or a disproportion between these elements and sodium, calcium and magnesium, might well account for the production of beriberi. As in the case of rickets and scurvy, because of the intricacy of the problems involved, and the large part played by the selective powers of the body

[12] Physiologische Chemie. 3. ed. (1894), 108–116.
[13] *Am. Journ. Phys.* (1900), 3, 327.

tissues, we may never be able to express in terms of chemical equations the pathologic processes of this disease, but I believe we do know enough of its etiology to prevent its occurrence.

By a proper diet we hope to eradicate it from the native troops. Of course, predisposing causes, such as bad ventilation, overexertion, exposure, etc., must receive their share of attention. As a result of our studies, certain changes are to be instituted in the Filipino ration. The rice is to be reduced to 16 ounces and native No. 2 rice substituted for the polished Siam grain now supplied. One and six-tenths ounces of beans are to be added. It is hoped that by these changes beriberi will disappear from the Scouts.

DISCUSSIONS ON THE PAPERS BY DOCTORS DE HAAN, FRASER, HIGHET, ARON, SHIBAYAMA, AND KILBOURNE.

Dr. Gilbert E. Brooke, port health officer, delegate from the Straits Settlements, Singapore.—I assure every one that I am very glad to have heard this series of papers, which shows how much careful work has been performed on this subject. That the removal of the outer covering of the rice grain is one of the causes of beriberi seems, from what we have heard, to be probable, the phosphorus content being reduced. However, I must say that it appears to me that we must be careful not to jump too hastily at conclusions. Before we can accept such a conclusion as this we should inquire into many other factors, for one, concerning the occurrence of beriberi in countries in which the principal diet is not rice, and another, its nonappearance in countries where rice does form the principal food. Another question is concerning the phosphorus content of other foods, which counterbalances the lack of phosphorus in white rice. The natives of this part of the world use fish wherever it is obtainable and fish contains phosphorus. Doctor Kilbourne, in his paper, states that beriberi was present in the Philippine Islands in 1882, but it seems unreasonable to assume that the natives at that time were already eating polished rice. They probably had the same diet as at present. Doctor Highet said that previous to 1890 white rice was quite expensive in Siam. I should like to ask him whether steam-milled rice had been exported from that country.

I have looked over the records for thirty years, covering the admission of cases of beriberi into the hospitals of the Straits Settlements and these develop the fact that no cases of the disease, or at least very few, occurred before 1878. Beriberi began to appear about the year 1880 and afterwards increased. We have had a considerable number of cases of beriberi annually from 1879 up to the present time. We began to use steam-milled rice in Singapore in 1890. There was no beriberi in Singapore jail from 1896 to 1897. The prisoners were given rice found in the market, which is the food of the native population, and the latter also had beriberi.

I had an opportunity of seeing the beginning of a case of beriberi taken from the quarantine station in Singapore. We had a number of hospital attendants, Chinese, who lived in barracks. They were doing the work of nurses. In October, 1896, we lacked one man. We secured a person about 31 years of age. He lived with the other coolies. They

had separate rooms, but they all ate the same food. Until December 25 none of these assistants went to Singapore. We allowed very little visiting, but permitted them to go once a month for a period of twenty-four hours. On December 25 this new man had 24 hours' leave and went to Singapore, but he returned the following morning. On January 5 this man complained of shortness of breath. I found him suffering from constriction of the chest; the following day his legs developed signs of weakness, on the seventh day he was very weak and on the eighth day he could no longer stand. I inquired concerning him on March 4 in order to discover what he had eaten during the twenty-four hours when he was in Singapore. I found that he had eaten two meals. I visited the house where he had eaten and found no beriberi there.

I bring this case to your attention so as to turn the latter on the possible origin of beriberi in this instance. There would seem to have been a short incubation period.

Dr. J. de Haan, director of the Government Medical Laboratory at Weltevreden, delegate from Her Majesty's Government of the Netherlands Indies.—Can Doctor Aron explain, by his view of the extraction of phytin, why the curative effect of *katjang idjo* is lost when the beans are heated to 120°?

Dr. Aldo Castellani, professor of tropical medicine and lecturer on dermatology, Ceylon Medical College, Colombo, delegate from the Government of Ceylon.—It was with great pleasure that I listened to these papers and to their discussion. I have always been strongly of the opinion that this matter should for the greater part be left to those who have especially worked with it; I have a certain amount of clinical experience with the disease; but have never undertaken any experimental work on it. The investigations of Doctors Fraser, Aron, and Kilbourne are, to my mind, the most convincing of all that I am aware of. In fact, I think that the supporters of the old theory of beriberi are in a very poor position; but I do not believe that the rice theory alone holds. I certainly agree with Doctor Aron that an insufficiently nutritrious diet may produce general debility, but may it not be that white rice and the general debility caused by its excessive consumption may render the individual more liable to be infected by the specific parasite causing beriberi? It seems to me that the rice theory does not explain the occurrence of the disease in some tropical countries and its absence in others. In Ceylon there are half a million Indian coolies who eat the same rice as those in India. In fact, the rice for our coolies is for the greater part imported from India; still, not a single case of beriberi has occurred in Ceylon.

In conclusion, I would ask to be allowed to express my admiration of the great work of Doctors Aron, Fraser, and Kilbourne.

Dr. Hans Aron, associate professor of physiology, Philippine Medical School.—In reply to Doctor de Haan's question, I will state that, in my

opinion, it is probable that if by a process of sterilization the soluble phosphorus compounds and phytin from beans (such as *mongo*) or from rice are extracted, this fact would explain the deleterious action of, say, sterilized beans, etc. I did not venture to touch upon this subject in my paper because I did not have sufficient experimental data, but this explanation is very clear to anyone who has worked with the question. An important point in regard to the process of sterilization would be to discover if, by this means, any extracts are produced which are separated from the beans and lost. A recent paper from the Physiological-Chemical Institute in Strassburg demonstrates that bread, when extracted, can not sustain mice in normal health, but if the extracts are added to the extracted bread, or unextracted bread is fed to the animals, they remain in good health.

In reply to Doctor Castellani's question, I would say that it can not be denied that the deleterious action of white rice could be explained by the supposition that a diet of this variety might favor the development of certain infective organisms. I can not agree as to the validity of calculations such as those made by Doctor Kilbourne. In the first place, even under military discipline, it is not possible to control the amount which each individual actually takes when the amount given to, say, 100 men only is known. I have observed that stricter rules are in force in Bilibid Prison than in the United States Army, and that even the prisoners exchange certain portions of their food; thus one man will trade meat for rice; or milk for cigarettes, and so on. I observed the same thing in Culion, when on a trip with Doctor Heiser. *Mongo* and rice are issued in Culion, a certain number of pounds for a certain number of men, but one man might eat rice alone, because he did not like *mongo;* others would take *mongo* because they were afraid of an attack of beriberi which at that time was prevalent in the colony.

In the second place, errors would be made in calculating the composition of foodstuffs according to tables or text-books. My own analyses demonstrate the great differences in the composition of two classes of the same food-stuffs, especially of rice. I do not know how anyone can determine the phosphorus content from the appearance of the rice alone. It is for these reasons that I doubt the value of such calculations, fraught as they are with two such grave errors.

Dr. Isaac W. Brewer, Medical Reserve Corps, United States Army.—Major Ruffner, United States Army, has asked me to present the statistics from Camp Connell, which during the past year has been one of the largest stations for native troops in the Islands. One hundred and eight cases occurred at that post from May to October, 1909. The disease was eliminated by increasing the amount of beef, bread, and beans given in the rations, and by limiting the rice to one meal daily,

which was taken at night. The entire command was examined every second Sunday and all suspicious cases were taken into the hospital so that the statistics are complete. The quality of the rice was not changed during this period.

Doctor de Haan.—I wish to reply to Doctor Castellani's objection to the view that the etiology of beriberi is due to the rice consumed, by the statement which I mentioned in my paper, namely, that neither in the blood nor the organs of men suffering from *Polyneuritis epidemica* nor in the organs of those who have died of this disease nor in the blood or organs of fowls suffering from experimental polyneuritis have antibodies or antigen been found. It is therefore improbable that an infection by microbes is the cause of beriberi, since we know that in all such infections antigen or antibodies are produced as a result of reaction by the infected organisms.

Dr. Gorosaku Shibayama, Institute for Infectious Diseases, Tokyo, delegate from His Imperial Japanese Majesty's Government.—It has been known for ten years that fowls, exclusively fed on polished rice, may contract polyneuritis, whereas the birds given the unpolished grain remain free from the disease. On the other hand, unpolished rice can produce polyneuritis when it has been heated to 120°. There is no chemical difference, especially in regard to the content of phosphorus, between heated and nonheated rice.

Furthermore, the occurrence of epidemics of beriberi in fishing villages has taught us that the inhabitants of the latter eat large quantities of fresh fish, and this diet contains, relatively, a large amount of phosphorus. We have treated many cases of beriberi with phytin, but we could not observe any very favorable results.

I would also, in this place, wish especially to emphasize the fact that the polyneuritis of fowls is not identical with beriberi, and that the experimental results obtained with these birds can not directly be interpreted in the same sense with human beings. So, for example, polyneuritis accompanies general cachexia and inanition in fowls, whereas beriberi, especially the acute, pernicious form, generally attacks well-nourished, muscular men.

Dr. Henry Fraser, director of the Institute for Medical Research, Kuala Lumpur, Federated Malay States, delegate from the Government of the Federated Malay States.—With reference to the observations of Major Kilbourne on the nutritive value of the diets issued to the people under his care, I do not think that we can estimate these from the composition of the various foodstuffs recorded in text-books because, in our experience, these records show considerable differences from the results of analyses carried out by us.

There are numerous species and varieties of rice; these differ in composition. Beef in the Tropics is poorer in fat and pork is richer in

fat than the corresponding articles as generally met with in Europe. If, then, we are to derive any information from the composition of the diets in respect to proteins, fats, carbohydrates and salts, actual analyses of the foodstuffs as issued must be made.

In our work at Durien Tipus we analyzed all the foodstuffs issued, and on comparing the diet issued to the party on parboiled rice with that issued to the party on white rice, no really important differences were observed, and both diets, considered in this way, should have sufficed for the physiological requirements of the individual.

We believe in our work at Durien Tipus that we excluded the operation of every other factor save rice in the production of beriberi.

Comparison of the composition of parboiled and white rices showed differences in respect to fat and ash, but the difference in amount of fats could not account for the results if we consider these bodies merely as the esters of fatty acids. The difference in amount of ash we have shown to depend mainly on the phosphorus compounds. Further investigation is necessary to explain the significance of this.

I believe that the method of estimating diets from the amount of proteins, fats, carbohydrates and ash contained in them will require reconsideration and in all probability readjustment.

In the light of recent research this method has shown itself to be crude and incapable of showing differences which may be of vital importance to the physiological requirements of the individual. Doctor Aron's researches are of the greatest importance and shed a large amount of light on this difficult problem, but I doubt if the difference in respect of phytin will explain the results.

In the case of the parboiled and white rices used by us and both derived from the same kind of paddy, we have estimated the phosphorus pentoxide in the washed and dried rices, because the rices are washed previous to cooking.

Now if we consider all the phosphorus estimated in this way to be combined as phytin and that a man receives one and a third pounds of rice daily, we find that the men on white rice would receive about 1.5 grams of phytin daily and those on parboiled rice about 3 grams of this substance.

The matter is, however, one of scientific interest and at present of no practical importance to those engaged in the prevention of beriberi.

Our researches have conclusively shown that beriberi can be prevented by the use of unpolished rice and as surely produced by the use of highly polished rice.

We have furnished you with three methods by which it is possible to determine the liability or otherwise of a given rice to produce beriberi.

1. Chemical.
2. Histological.

3. By feeding experiments on fowls.

Of course, if the dietary of those peoples among whom beriberi is occurring be improved, the use of highly polished rice might be continued and no untoward effect occur, but so long as these peoples continue to partake of a diet in which rice constitutes the staple, our efforts must be directed to maintaining the rice at a standard sufficient for their physiological requirements.

I wish to correct here one misapprehension. It is not the removal of the pericarp which makes the rice harmful, but of the layers subjacent to the pericarp (subpericarpal layers); the cells composing those layers differ in respect of their constituents from the cells composing the central part of the endosperm.

Dr. H. Campbell Highet, principal medical officer local government, Bangkok, delegate from His Imperial Majesty's Government, Siam.—In discussing epidemics of beriberi in mines, the theory that arsenical poisoning may be the cause of the particular polyneuritis observed must not be forgotten. I can give no information with respect to how long white Siam rice had been imported into Singapore. As regards the one case of beriberi which arose amongst seven members of Doctor Brooke's quarantine staff, it should always be borne in mind that one swallow does not make a summer, and that this one case proved nothing. However, allowing that this case was really beriberi and that the symptoms appeared to develop after nearly three months' residence on the quarantine island, this was not remarkable. If we were to speak of such a thing as an incubation period, I could not fix any definite time for such a period. It may vary from a few days to months or even to a year or more. Why may Doctor Brookes's case not have been a relapse? Men with extended experience in beriberi know that before starting upon feeding experiments the investigator must be careful in the examination of the persons about to undergo treatment in order to eliminate old or incipient cases of the disease. My own experience is that beriberi once contracted is most difficult to eradicate entirely, and that months or even years afterwards some sudden strain upon the health might bring about a condition in every way like acute beriberi. A case of a Siamese police sergeant may be cited, who, after a prolonged residence in hospital, suffering from beriberi, returned to duty and did good work for eighteen months. Having had a dispute with one of his subordinates, he was put into a police cell to await trial. In thirty-six hours he became so ill that he was transferred to hospital, where he died in a few hours, with all the symptoms of acute, wet beriberi. On post-mortem examination, it was quite evident that the lesions of the heart were the result of prolonged illness. He had the largest "bullock's heart" I ever saw. His previous history was not known, but the conclusion might easily have been reached

that this was a case of acute, wet beriberi whereas it was simply a relapse in a very chronic case.

Dr. H. M. Neeb, medical officer of the first class, delegate from Her Majesty's Government of the Netherlands Indies.—I wish to ask Doctor Shibayama if, in his investigations of the cases observed in the Island of Banca, in the mines, where fresh rice was given, he made sure that there were not previous cases or relapses. It is true that patients who have had beriberi and recovered therefrom, so as apparently to be in a healthy condition, a long time afterwards if suddenly placed in bad surroundings, for instance, violent change of climate or excessive hard labor, will not infrequently suffer a relapse. If, now, patients are given fresh, unpolished rice, it might appear, if they contract beriberi, as if this food were the cause, whereas there may be nothing but a relapse produced by other conditions and not by the rice.

In regard to the question of beriberi on sailing ships, I wish to state that about a year ago Professor Nocht studied two cases where men had died from this cause. Doctor Rodenwaldt examined pathologic-anatomic preparations of the spinal cord and peripheral nerves and found the changes to be quite the same as those which occur in exotic beriberi, so that Professor Nocht has come to the opinion that both varieties of beriberi are identical.

Kakke (beriberi) has been termed *Polyneuritis epidemica* during this discussion, but investigations in Japan have demonstrated, to a certain extent, that the alterations in the nerves are mainly secondary degenerations, the principal change being in the vascular system. For that reason the symptom complex differentiating *kakke* (beriberi) and intoxication polyneuritis is easy to determine. I do not wish to confuse beriberi with the polyneuritis of intoxication.

Dr. E. D. Kilbourne, captain, medical corps United States Army, member of the United States Army Board for the Study of Tropical Diseases as they exist in the Philippine Islands.—In reply to Doctors Aron and Fraser I will state that the amounts of phosphorus and potassium shown in the table were average amounts taken from a large number of men over a period of several months. Although the figures may not be absolutely correct, they are of value in showing the relative amounts of phosphorus and potassium in the three groups, and show a greater reduction in potassium than phosphorus in the affected groups. The phosphorus and potassium contents of various foods used to compute the tables were themselves average figures taken from a number of different analyses, so the amounts given in the table are considered to be fairly correct. I fully appreciate the criticism which Doctor Aron makes of my calculations, but analyses of identical rice made in different laboratories show at least as great variations in composition as he states occur between different kinds of rice.

Dr. Francis Clark, medical officer of health, delegate from the Government of Hongkong.—In order to strengthen the hands of medical officers concerned in public health administration or in the medical supervision of native laborers, and after consultation with some of those attending this congress, I desire, after the discussion is closed, to move a resolution in regard to the formation of a committee on beriberi.

The wording is immaterial so long as you are prepared to confirm the principal involved, namely, that this disease is occasioned by the consumption of white or highly polished rice as the staple article of diet. The adoption of such a resolution will yield a practical issue to the discussion and will enable us to take early steps to protect the natives under our care from a disease which is responsible for much suffering and many deaths.[1]

[1] The following resolution was finally passed by the association (Ed.):

"*Resolved*, That in the opinion of this Association sufficient evidence has now been produced in support of the view that beriberi is associated with the continuous consumption of white (polished) rice as the staple article of diet, and the Association accordingly desires to bring this matter to the notice of the various Governments concerned."

REVIEW.

Retinitis Pigmentosa. With an Analysis of Seventeen Cases in Deaf-Mutes.—By William T. Shoemaker, M. D. Laboratory Examinations of the Blood and Urine, by John M. Swan, M. D. Cloth. Pp. IV+106. Price $2. 1st ed. Philadelphia: J. B. Lippincott Company.

This little book on retinitis pigmentosa gives an exceedingly satisfactory discussion of the subject. The description of the pathology, symptoms and fundus changes are concise and yet complete. The plates are beautiful and anyone able to view the fundus should, with their aid, have no trouble in making a diagnosis even in a first case.

The author's statement that the disease is particularly prevalent in the Orient should interest practitioners in this region and induce them to investigate closely any complaint of night-blindness by patients. This symptom with contraction of the visual fields and the retinal pigmentation make the disease "one of the most easily recognized in the domain of ophthalmology."

Doctor Shoemaker's conclusions may be briefly summarized as follows:

1. That the disease is a degeneration, not an inflammation, of the entire neurovascular tract of the peripheral end-organ of vision.

2. That the retinal changes are secondary to changes in the choroid.

3. That the disease is congenital in all cases, however long the symptoms may be delayed, and that it may be considered as a stigma of degeneracy.

4. That heredity is a potent etiologic factor; parental consanguinity is of importance only in connection with heredity and environment; maternal impressions can not be excluded as a possible cause; and syphilis as a cause of true retinitis pigmentosa is not established.

5. That the distribution is of more importance than the mere presence of retinal pigment in making the diagnosis.

6. That the disease is *always* bilateral.

R. P. O'C.

NOTICE.

Beginning with the January, 1910, issue the old-established Medical Review of Reviews will be edited by Dr. William J. Robinson, editor and founder of the famous Critic and Guide, Therapeutic Medicine, and The American Journal of Urology.

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